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PUTREFACTION AND INFECTION

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ESSAYS ON THE

FLOATING-MATTER OF THE AIR

IN RELATION TO

PUTREFACTION AND INFECTION

BY

JOHN TYNDALL, F.R.S.

(M.D. TÜBINGEN)

LONDON
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INTRODUCTORY NOTE.

THE VIRTUAL TRIUMPH of the Antiseptic system of surgery, based as that system is on the recognition of *living contagia* as the agents of putrefaction, is of good augury as regards the receptivity of the public mind to new views respecting the nature of contagia generally.

To the credit of English surgeons it stands recorded that, guided by their practical sagacity, they had adopted in their hospitals measures of amelioration which reduced, almost to a minimum, the rate of mortality arising from the 'mortification' of wounds. They had discovered the evils incident to 'dirt;' and, by keeping dirt far away from them, they had saved innumerable lives, which would undoubtedly have succumbed under conditions prevalent in many of the hospitals of continental Europe.

In thus acting, English surgeons were, for the most part, 'wiser than they knew.' Their knowledge, however momentous in its practical applications, was still empirical knowledge. That dirt was fatal they had discovered; but why it was fatal few of them knew.

At this point Lister came forward with a scientific principle which rendered all plain. Dirt was fatal, not as dirt, but because it contained living germs which, as Schwann was the first to prove, are the cause of putrefaction. Lister extended the generalization of Schwann from dead matter to living matter, and by this apparently simple step revolutionized the art of surgery. He changed it, in fact, from an art into a science.

'Listerism' is sometimes spoken of as if it merely consisted in the application of carbolic acid spray; but no man of any breadth of vision will regard the subject thus. The antiseptic system had been enunciated, expounded, and illustrated, prior to the introduction of the spray. The spray is a mere offshoot of the system—elegant and effective it is true, but still a matter of detail. In company with my excellent friend Mr. John Simon, I once visited St. Bartholomew's Hospital, and became acquainted, in its wards, with the practice of the late Mr. Callender. The antiseptic system was there as stringently applied as at King's College. Immediately before his departure to America I spoke to Mr. Callender on this subject; and he then told me expressly, that his aim and hope had been, not to introduce a new principle, but to simplify the methods of Lister. And yet Mr. Callender's practice is sometimes spoken of as if it were, in principle, different from that of his eminent contemporary.

It is interesting, and indeed pathetic, to observe how long a discovery of priceless value to humanity

may be hidden away, or rather lie openly revealed, before the final and apparently obvious step is taken towards its practical application. In 1837, Schwann clearly established the connexion between putrefaction and microscopic life; but thirty years had to elapse before Lister extended to wounds the researches of Schwann on dead flesh and animal infusions. Prior to Lister the possibility of some such extension had occurred to other minds. Penetrative men had seen that the germs which produce the putrefaction of meat might also act with fatal effect in the wards of a hospital.

Thus, for example, in a paper read before the British Medical Association at Cambridge in 1864, Mr. Spencer Wells pointed out that the experiments of Pasteur, then recent, had 'all a very important bearing upon the development of purulent infection, and the whole class of diseases most fatal in hospitals and other overcrowded places.' Mr. Wells did not, as far as I know, introduce any systematic mode of combating the organisms whose power he so early recognised. But, I believe, in hardly any other department of surgery has the success of the antiseptic system been more conspicuous and complete than in that particular sphere of practice in which Mr. Wells has won so great a name.

A remark in the paper just referred to would seem to indicate that, in regard to the further possible influence of germs, the thoughts of Mr. Spencer Wells had passed beyond the bounds of pure surgical practice. 'Their influence,' he says, 'on the propagation of

epidemic and contagious diseases has yet to be made out.'

This shows that at the time here referred to the Germ Theory, in its wider medical sense, had begun to ferment in England. Two years, indeed, prior to the above occasion, and for the use of the same Association as that addressed by Mr. Wells, the late Dr. William Budd had drawn up a series of 'Suggestions towards a Scheme for the Investigation of Epidemic and Epizootic Diseases,' which strikingly illustrate the insight of a man of genius, withdrawn from the stimulus of the metropolis, and working alone, at a time when the whole medical profession in England entertained views opposed to his. Budd states in succession, and with perfect clearness, the points which he considers most worthy of the attention of the Association. He recommends inquiry as to the nature of the evidence alleged to prove the disease under investigation to be contagious or communicable. Whether such disease admits of being artificially propagated by inoculation or otherwise. Through what surface or surfaces the virus may be shown to enter the body, and to leave it, when the disease is taken in the natural way. Whether the disease is distinguished by eruptions external or internal. Whether it has a period of true incubation; and if so, what are the length and limits of that period. Whether one attack, as in smallpox and many other contagious diseases, preserves against future attacks. Whether in the case of human disease animals as well as man are susceptible, and if so, what animals. What is the evidence, if any, as to the particular country or region in which the disease first appeared. What are

its present geographical limits. Whether there is any evidence of its modern or recent introduction into countries previously exempt. How far any such disease may have been prevented from invading new countries, or from spreading from any particular centre, by measures directed against contagion. Above all, to determine what is the nature, and what the true value, of the evidence supposed to show that the specific poison of a contagious disease may originate spontaneously, or be generated *de novo*. 'What we most want to know,' adds Budd, 'in regard to this whole group of diseases is, *where, and how, the specific poisons which cause them, breed and multiply.*'

Budd's own relation to the question here raised was distinct and, under the circumstances, impressive. 'After giving many years of time and thought to an examination of the evidence bearing on this question,' he comes to the conclusion 'that there is no proof whatever' that the poisons of specific contagious diseases ever originate spontaneously. 'That the evidence on which the contrary conclusion is founded is negative only; that evidence of precisely the same order, only to all appearance still more cogent, would prove animals and plants, even of large species, to originate spontaneously; that this evidence is therefore of no weight; and, lastly, that all the really important facts point the other way, and tend to prove that these poisons (to use a term which is probably provisional only), like animals and plants, however they may have once originated, are only propagated now by the law of continuous succession.'

The word 'poisons,' here provisionally employed, was a concession on Budd's part to his weaker brethren; for he, without a shade of doubt, considered the poison to be a real living *seed*. There was, I believe, but one physician of eminence in England who, at the time here referred to, shared this conviction, and who imparted to Budd the incalculable force derived from the approbation and encouragement of a wise and celebrated man. It gives me singular pleasure to write down here the name of the venerable Sir Thomas Watson, who lent to William Budd unfailing countenance and support, and who has lived to see that the views which commended themselves to his philosophic judgment are at the present moment advancing with resistless momentum among the members of the medical profession. It was far otherwise at the time to which we here refer. 'Opinions like these,' said Budd, 'are no doubt, at present, those of a small minority. A very large, and by far the most influential school in this country—a school which probably embraces the great majority of medical practitioners, and the whole of the "sanitary public"—holds the exact contrary; and teaches that sundry of these poisons are constantly being generated *de novo* by the material conditions which surround us.'

Budd's remark regarding the spontaneous generation of 'animals and plants, even of large species,' is both pregnant and pertinent. In reference to special and solitary outbreaks of contagious fever, I have frequently heard physicians of distinction affirm, without apparent misgiving, the 'impossibility' of importation from

without. On such occasions a reply, in the strict sense affirmed by William Budd, was always at hand; for I was able to adduce cases of solitary mushrooms, found upon out of the way Alpine slopes, to which the evidence would apply with greater force than to the cases on which the physicians referred to based their conclusions. With the atmosphere as a vehicle of universal intercommunication, it is hard to see any just warrant for the reliance of medical men upon the negative evidence stigmatized by Budd as valueless. It is, however, evidence by which many physicians are still influenced, and the effects of which it will probably require a generation of doctors, brought up under other conditions of culture and of practice, to wholly sweep away.

These conditions are growing up around us, and their influence will be all-pervading before long. Never before was medicine manned and officered as it is now. To name here the workers at present engaged in the investigation of communicable diseases would be to extend beyond all reasonable limits this Introductory Note. On the old Baconian lines of observation and experiment the work is carried on. The intercommunication of scientific thought plays here a most important part. It will probably have been noticed, that while physiologists and physicians in England and elsewhere were drawing copiously from the store of facts furnished by the researches of Pasteur, that admirable investigator long kept himself clear of physiology and medicine. There is, indeed, reason to believe that he was spurred on to his most recent achievements by the papers of

Burdon Sanderson, Koch, and others. The union of scientific minds is, or ought to be, organic. They are parts of the same body, in which every member, under penalty of atrophy and decay, must discharge its due share of the duty imposed upon the whole. Of this 'body,' a short time since, England provided one of the healthiest limbs ; but round that limb legislation has lately thrown a ligature, which threatens to damage its circulation and to divert its energies into foreign channels. In observational medicine one fine piece of work may be here referred to—the masterly inquiry of Dr. Thorne Thorne into the outbreak of typhoid fever at Caterham and Redhill. Hundreds were smitten by this epidemic, and many died. The qualities of mind illustrated in Dr. Thorne's inquiry match those displayed by William Budd in his memorable investigation of a similar outbreak in Devonshire. Dr. Budd's process was centrifugal—tracing from a single case in the village of North Tawton, the ravages of the fever far and wide. Dr. Thorne's process was centripetal—tracing the epidemic backwards from the multitude of cases first presented, to the single individual whose infected excreta, poured into the well at Caterham, were the cause of all.

The Essays here presented to the reader belong to the A B C of the great subject touched upon in the foregoing Note. The two principal ones, namely, Essays II. and III., were prepared for the Royal Society, and are published in the 'Philosophical Transactions' for 1876 and 1877. But, though written for

that learned body, I sought to render their style and logic so clear as to render them accessible to any fairly cultivated mind. The Essays on 'Fermentation' and 'Spontaneous Generation' have already appeared elsewhere; while the first Essay, on 'Dust and Disease,' has been for some years before the public. It may be regarded as a kind of popular introduction to the more strenuous and original labours which follow it.

The Essay most likely to try the reader's patience is No. III. On the whole, however, and particularly in its bearings on the Germ Theory of disease, it is probably the most important of all. The difficulties which sometimes beset the experimenter in these investigations are best illustrated by this Essay. It shows, to my mind in a very impressive manner, the analogy of the spread of infection among organic infusions with its mode of propagation among human beings. The vital resistance of certain germs to heat is strikingly illustrated in the third Essay, one infusion being there proved to maintain its potentiality of life intact after eight hours' continuous exposure to the temperature of boiling water. Under the plain guidance of the Germ Theory, it is however shown that an infusion of this stubborn character may be infallibly sterilized by discontinuous heating, in one hundredth part of the time requisite when the boiling is continuous. Another question, to my mind of fundamental importance, is also disposed of in Essay III., where it is shown that the germs which exhibited the foregoing resistance are neither contained in the air, nor attached to the surface of the vessel, above the liquid, but that

they manifest their extraordinary vitality in the body of the liquid itself.

On public sympathy the sanitary physician has mainly to rely for support, in a country where sanitary matters are left so much in the hands of the public itself as they are in England. But sympathy without cause—that is to say, without some basis of knowledge—is hardly to be expected. It is as a contribution to such knowledge that these Essays have been collected, and thrown into their present handy form.

J. Tyndall.

Royal Institution:
August 1881.

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ON DUST AND DISEASE.

I.

Experiments on Dusty Air.

Solar light, in passing through a dark room, reveals its track by illuminating the dust floating in the air. 'The sun,' says Daniel Culverwell, 'discovers atomes, though they be invisible by candle-light, and makes them dance naked in his beams.'

In my researches on the decomposition of vapours by light, I was compelled to remove these 'atomes' and this dust. It was essential that the space containing the vapours should embrace no visible thing—that no substance capable of scattering light in the slightest sensible degree should, at the outset of an experiment, be found in the wide 'experimental tube' in which the vapour was enclosed.

For a long time I was troubled by the appearance there of floating matter, which, though invisible in diffuse daylight, was at once revealed by a powerfully condensed beam. Two U-tubes were placed in succession in the path of the air, before it entered the liquid whose vapour was to be carried into the experimental tube. One of the U-tubes contained fragments of marble wetted with a strong solution of caustic potash;

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the other, fragments of glass wetted with concentrated sulphuric acid which, while yielding no vapour of its own, powerfully absorbs the aqueous vapour of the air. To my astonishment, the air of the Royal Institution, sent through these tubes at a rate sufficiently slow to dry it, and to remove its carbonic acid, carried into the experimental tube a considerable amount of mechanically suspended matter, which was illuminated when the beam passed through the tube. The effect was substantially the same when the air was permitted to bubble through the liquid acid, and through the solution of potash.

I tried to intercept this floating matter in various ways; and on October 5, 1868, prior to sending the air through the drying apparatus, it was carefully permitted to pass over the tip of a spirit-lamp flame. The floating matter then no longer appeared, having been burnt up by the flame. It was therefore *organic matter*. I was by no means prepared for this result; having previously thought that the dust of our air was, in great part, inorganic and non-combustible.[1]

I had constructed a small gas-furnace, now much employed by chemists, containing a platinum tube, which could be heated to vivid redness.[2] Within this

[1] According to an analysis kindly furnished to me by Dr. Percy, the dust collected *from the walls* of the British Museum contains fully 50 per cent. of inorganic matter. I have every confidence in the results of this distinguished chemist; they show that the *floating* dust of our rooms is, as it were, winnowed from the heavier matter. As bearing directly upon this point I may quote the following passage from Pasteur: 'Mais ici se présente une remarque: la poussière que l'on trouve à la surface de tous les corps est soumise constamment à des courants d'air, qui doivent soulever les particules les plus légères, au nombre desquelles se trouvent, sans doute, de préférence les corpuscules organisés, œufs ou spores, moins lourds généralement que les particules minérales.'

[2] Pasteur was, I believe, the first to employ such a tube.

tube was a roll of platinum gauze, which, while it permitted the air to pass through it, ensured the practical contact of the dust with the incandescent metal. The air of the laboratory was permitted to enter the experimental tube, sometimes through the cold, and sometimes through the heated, tube of platinum. In the first column of the following fragment of a long table of results, the quantity of air operated on is expressed by the depression of the mercury gauge of the air-pump. In the second column the condition of the platinum tube is mentioned, and in the third the state of the air in the experimental tube.

Quantity of air	State of platinum tube	State of experimental tube
15 inches . .	Cold . .	Full of particles.
30 „ . .	Red-hot . .	Optically empty.

The phrase 'optically empty' shows that when the conditions of perfect combustion were present, the floating matter totally disappeared.

In a cylindrical beam, which strongly illuminated the dust of the laboratory, I placed an ignited spirit-lamp. Mingling with the flame, and round its rim, were seen curious wreaths of darkness resembling an intensely black smoke. On placing the flame at some distance below the beam, the same dark masses stormed upwards. They were blacker than the blackest smoke ever seen issuing from the funnel of a steamer; and their resemblance to smoke was so perfect as to lead the most practised observer to conclude that the apparently pure flame of the alcohol lamp required but a beam of sufficient intensity to reveal its clouds of liberated carbon.

But is the blackness smoke? This question presented itself in a moment and was thus answered: A red-hot poker was placed underneath the beam: from

it the black wreaths also ascended. A large hydrogen flame was next employed, and it produced those whirling masses of darkness, far more copiously than either the spirit-flame or poker. Smoke was therefore out of the question.[1]

What, then, was the blackness? It was simply that of stellar space; that is to say, blackness resulting from the absence from the track of the beam of all matter competent to scatter its light. When the flame was placed below the beam the floating matter was destroyed *in situ*; and the air, freed from this matter, rose into the beam, jostled aside the illuminated particles, and substituted for their light the darkness due to its own perfect transparency. Nothing could more forcibly illustrate the invisibility of the agent which renders all things visible. The beam crossed, unseen, the black chasm formed by the transparent air, while, at both sides of the gap, the thick-strewn particles shone out like a luminous solid under the powerful illumination.

It is not, however, necessary to burn the particles to produce a stream of darkness. Without actual combustion, currents may be generated which shall displace the floating matter, and appear dark amid the surrounding brightness. I noticed this effect first on placing a red-hot copper ball below the beam, and permitting it to remain there until its temperature had fallen below that of boiling water. The dark currents, though much enfeebled, were still produced. They may also be produced by a flask filled with hot water.

[1] In none of the public rooms of the United States where I had the honour to lecture was this experiment made. The organic dust was too scanty. Certain rooms in England—the Brighton Pavilion, for example—also lack the necessary conditions.

To study this effect a platinum wire was stretched transversely under the beam, the two ends of the wire being connected with the two poles of a voltaic battery. To regulate the strength of the current a rheostat was placed in the circuit. Beginning with a feeble current the temperature of the wire was gradually augmented; but long before it reached the heat of ignition, a flat stream of air rose from it, which when looked at edgeways appeared darker and sharper than one of the blackest lines of Fraunhofer in the purified spectrum. Right and left of this dark vertical band the floating matter rose upwards, bounding definitely the non-luminous stream of air. What is the explanation? Simply this: The hot wire rarefied the air in contact with it, but it did not equally lighten the floating matter. The convection current of pure air therefore passed upwards among the inert particles, dragging them after it right and left, but forming between them an impassable black partition. This elementary experiment enables us to render an account of the dark currents produced by bodies at a temperature below that of combustion.

When the platinum wire is intensely heated, the floating matter is not only displaced, but destroyed. I stretched a wire about 4 inches long through the air of an ordinary glass shade resting on cotton-wool, which also surrounded the rim. The wire being raised to a white heat by an electric current, the air expanded, and some of it was forced through the cotton-wool. When the current was interrupted, and the air within the shade cooled, the returning air did not carry motes along with it, being filtered by the wool. At the beginning of this experiment the shade was charged with floating matter; at the end of half an hour it was optically empty.

On the wooden base of a cubical glass shade, a cubic foot in volume, upright supports were fixed, and from one support to the other 38 inches of platinum wire were stretched in four parallel lines. The ends of the platinum wire were soldered to two stout copper wires which passed through the base of the shade and could be connected with a battery. As in the last experiment the shade rested upon cotton-wool. A beam sent through the shade revealed the suspended matter. The platinum wire was then raised to whiteness. In five minutes there was a sensible diminution of the matter, and in ten minutes it was totally consumed.

Oxygen, hydrogen, nitrogen, carbonic acid, so prepared as to exclude all floating particles, produce, when poured or blown into the beam, the darkness of stellar space. Coal-gas does the same. An ordinary glass shade, placed in the air with its mouth downwards, permits the track of the beam to be seen crossing it. When coal-gas or hydrogen is allowed to enter the shade by a tube reaching to its top, the gas gradually fills the shade from above downwards. As soon as it occupies the space crossed by the beam, the luminous track is abolished. Lifting the shade so as to bring the common boundary of gas and air above the beam, the track flashes forth. After the shade is full, if it be inverted, the pure gas passes upwards like a black smoke among the illuminated particles.

The Germ Theory of Contagious Disease.

There is no respite to our contact with the floating matter of the air. We not only suffer from its mechanical irritation, but it is a growing belief that a portion of it lies at the root of a class of disorders

most deadly to man. And what is this portion? It was some time ago the current belief that epidemic diseases generally were propagated by a kind of malaria, which consisted of organic matter in a state of motor-decay; that when such matter was taken into the body through the lungs, skin, or stomach, it had the power of spreading there the destroying process by which itself had been assailed. Such a power, it was alleged, was visibly exerted in the case of yeast. A little leaven was seen to leaven the whole lump—a mere speck of matter, in this supposed state of decomposition, being apparently competent to propagate indefinitely its own decay. Why should not a bit of rotten malaria within the human body act in a similar way? In 1836 a very unexpected reply was given to this question. In that year Cagniard de la Tour discovered the *yeast-plant*—a living organism, which when placed in a proper medium feeds, grows, and reproduces itself, and in this way carries on the process which we name fermentation. Here we have active life instead of motor-decay. By this discovery fermentation was connected with organic growth.

Schwann, of Berlin, discovered the yeast-plant independently about the same time; and in February, 1837, he also announced the important result, that when a decoction of meat is effectually screened from ordinary air, and supplied solely with calcined air, putrefaction never sets in. Putrefaction, therefore, he affirmed to be caused, not by the air, but by something in the air which could be destroyed by a sufficiently high temperature. The results of Schwann were confirmed by the independent experiments of Helmholtz, Ure, and Pasteur, while other methods, pursued by Schultze, and by Schroeder and Dusch, led to the same result. But as regards fermentation, the minds of chemists,

influenced probably by the great authority of Gay-Lussac, fell back upon the old notion of matter in a state of decay. It was not, they said, the living yeast-plant, but the dead or dying part of it, which, assailed by oxygen, produced the fermentation. But, as a matter of fact, when the plant is killed the ferment disappears. Mediate or immediate, the real 'ferments' are living organisms which find in fermentable substances their necessary food.

Side by side with these researches and discoveries, and fortified by them and others, has run the *germ theory* of epidemic disease. The notion was expressed by Kircher, and favoured by Linnæus, that epidemic diseases may be due to germs which float in the atmosphere, enter the body, and produce disturbance by the development within the body of parasitic life. The strength of this theory consists in the perfect parallelism of the phenomena of contagious disease with those of life. As a planted acorn gives birth to an oak, competent to produce a whole crop of acorns, each gifted with the power of reproducing its parent tree; and as thus from a single seedling a whole forest may spring; so, it is contended, these epidemic diseases literally plant their seeds, grow, and shake abroad new germs, which, meeting in the human body their proper food and temperature, finally take possession of whole populations. There is nothing to my knowledge in pure chemistry which resembles the power of propagation and self-multiplication possessed by the matter which produces epidemic disease. If you sow wheat you do not get barley; if you sow small-pox you do not get scarlet-fever, but small-pox indefinitely multiplied, and nothing else. The matter of each contagious disease reproduces itself as rigidly as if it were (as Miss Nightingale puts it) dog or cat.

Parasitic Diseases of Silkworms. Pasteur's Researches.

It is admitted on all hands that some diseases are the product of parasitic growth. Both in man and in lower creatures, the existence of such diseases has been demonstrated. I am enabled to lay before you an account of an epidemic of this kind, thoroughly investigated and successfully combated by M. Pasteur. For fifteen years a plague had raged among the silkworms of France. They had sickened and died in multitudes, while those that succeeded in spinning their cocoons furnished only a fraction of the normal quantity of silk. In 1853 the silk culture of France produced a revenue of one hundred and thirty millions of francs. During the twenty previous years the revenue had doubled itself, and no doubt was entertained as to its further augmentation. The weight of the cocoons produced in 1853 was 52,000,000 pounds; in 1865 it had fallen to 8,000,000, the fall entailing, in a single year, a loss of 100,000,000 francs.

The country chiefly smitten by this calamity happened to be that of the celebrated chemist Dumas, now perpetual secretary of the French Academy of Sciences. He turned to his friend, colleague, and pupil, Pasteur, and besought him, with an earnestness which the circumstances rendered almost personal, to undertake the investigation of the malady. Pasteur at this time had never seen a silkworm, and he urged his inexperience in reply to his friend. But Dumas knew too well the qualities needed for such an inquiry to accept Pasteur's reason for declining it. 'Je mets,' said he, 'un prix extrême à voir votre attention fixée sur la question qui intéresse mon pauvre pays; la misère surpasse tout ce

que vous pouvez imaginer.' Pamphlets about the plague had been showered upon the public, the monotony of waste paper being broken, at rare intervals, by a more or less useful publication. 'The Pharmacopœia of the Silkworm,' wrote M. Cornalia in 1860, 'is now as complicated as that of man. Gases, liquids, and solids have been laid under contribution. From chlorine to sulphurous acid, from nitric acid to rum, from sugar to sulphate of quinine,—all has been invoked in behalf of this unhappy insect.' The helpless cultivators, moreover, welcomed with ready trustfulness every new remedy, if only pressed upon them with sufficient hardihood. It seemed impossible to diminish their blind confidence in their blind guides. In 1863 the French Minister of Agriculture signed an agreement to pay 500,000 francs for the use of a remedy, which its promoter declared to be infallible. It was tried in twelve different departments of France, and found perfectly useless. In no single instance was it successful. It was under these circumstances that M. Pasteur, yielding to the entreaties of his friend, betook himself to Alais in the beginning of June, 1865. As regards silk husbandry, this was the most important department in France, and it was the most sorely smitten by the plague.

The silkworm had been previously attacked by *muscardine*, a disease proved by Bassi to be caused by a vegetable parasite. This malady was propagated annually by the parasitic spores. Wafted by winds, they often sowed the disease in places far removed from the centre of infection. Muscardine is now said to be very rare, a deadlier malady having taken its place. This new disease is characterized by the black spots which cover the silkworms; hence the name *pébrine*, first applied to the plague by M. de Quatrefages,

and adopted by Pasteur. Pébrine declares itself in the stunted and unequal growth of the worms, in the languor of their movements, in their fastidiousness as regards food, and in their premature death. The course of discovery as regards the epidemic is this: In 1849 Guérin Méneville noticed in the blood of silkworms vibratory corpuscles, which he supposed from their motions to be endowed with independent life. Filippi, however, showed that the motion of the corpuscles was the well-known Brownian motion; but he committed the error of supposing the corpuscles to be normal to the life of the insect. Possessing the power of indefinite self-multiplication, they are really the cause of its mortality—the form and substance of its disease. This was well described by Cornalia; while Lebert and Frey subsequently found the corpuscles not only in the blood, but in all the tissues of the insect. Osimo, in 1857, discovered them in the eggs; and on this observation Vittadiani founded, in 1859, a practical method of distinguishing healthy from diseased eggs. The test often proved fallacious, and it was never extensively applied.

These living corpuscles first take possession of the intestinal canal, and spread thence throughout the body of the worm. They fill the silk cavities, the stricken insect often going automatically through the motions of spinning, without any material to work upon. Its organs, instead of being filled with the clear viscous liquid of the silk, are packed to distension by the corpuscles. On this feature of the plague Pasteur fixed his entire attention. The cycle of the silkworm's life is briefly this: From the fertile egg comes the little worm, which grows, and casts its skin. This process of moulting is repeated two or three times at intervals during the life of the insect. After

the last moulting the worm climbs the brambles placed to receive it, and spins among them its cocoon. It passes thus into a chrysalis; the chrysalis becomes a moth, and the moth, when liberated, lays the eggs which form the starting-point of a new cycle. Now Pasteur proved that the plague-corpuscles might be incipient in the egg, and escape detection; they might also be germinal in the worm, and still baffle the microscope. But as the worm grows, the corpuscles grow also, becoming larger and more defined. In the aged chrysalis they are more pronounced than in the worm; while in the moth, if either the egg or the worm from which it comes should have been at all stricken, the corpuscles infallibly appear, offering no difficulty of detection. This was the first great point made out in 1865 by Pasteur. The Italian naturalists, as aforesaid, recommended the examination of the eggs before risking their incubation. Pasteur showed that both eggs and worms might be smitten, and still pass muster, the culture of such eggs or such worms being sure to entail disaster. He made the moth his starting-point in seeking to regenerate the race.

Pasteur made his first communication on this subject to the Academy of Sciences in September, 1865. It raised a cloud of criticism. Here, forsooth, was a chemist rashly quitting his proper *métier* and presuming to lay down the law for the physician and biologist on a subject which was eminently theirs. 'On trouva étrange que je fusse si peu au courant de la question; on m'opposa des travaux qui avaient paru depuis longtemps en Italie, dont les résultats montraient l'inutilité de mes efforts, et l'impossibilité d'arriver à un résultat pratique dans la direction que je m'étais engagé. Que mon ignorance fut grande au sujet des recherches sans nombre qui avaient paru depuis quinze

années.' Pasteur heard the buzz, but he continued his work. In choosing the eggs intended for incubation, the cultivators selected those produced in the successful 'educations' of the year. But they could not understand the frequent and often disastrous failures of their selected eggs; for they did not know, and nobody prior to Pasteur was competent to tell them, that the finest cocoons may envelope doomed corpusculous moths. It was not, however, easy to make the cultivators accept new guidance. To strike their imagination, and if possible determine their practice, Pasteur hit upon the expedient of prophecy. In 1866 he inspected, at St. Hippolyte-du-Fort, fourteen different parcels of eggs intended for incubation. Having examined a sufficient number of the moths which produced these eggs, he wrote out the prediction of what would occur in 1867, and placed the prophecy as a sealed letter in the hands of the Mayor of St. Hippolyte.

In 1867 the cultivators communicated to the mayor their results. The letter of Pasteur was then opened and read, and it was found that in twelve out of fourteen cases there was absolute conformity between his prediction and the observed facts. Many of the groups had perished totally; the others had perished almost totally; and this was the prediction of Pasteur. In two out of the fourteen cases, instead of the prophesied destruction, half an average crop was obtained. Now, the parcels of eggs here referred to were considered healthy by their owners. They had been hatched and tended in the firm hope that the labour expended on them would prove remunerative. The application of the moth-test for a few minutes in 1866 would have saved the labour and averted the disappointment. Two additional parcels of eggs were at the same time submitted to Pasteur. He pronounced them healthy; and

his words were verified by the production of an excellent crop. Other cases of prophecy still more remarkable, because more circumstantial, are recorded in Pasteur's work on the Diseases of Silkworms.

Pasteur subjected the development of the corpuscles to a searching investigation, and followed out with admirable skill and completeness the various modes by which the plague was propagated. From moths perfectly free from corpuscles he obtained healthy worms, and selecting 10, 20, 30, 50, as the case might be, he introduced into the worms the corpusculous matter. It was first permitted to accompany the food. Let us take a single example out of many. Rubbing up a small corpusculous worm in water, he smeared the mixture over mulberry-leaves. Assuring himself that the leaves had been eaten, he watched the consequences from day to day. Side by side with the infected worms he reared their fellows, keeping them as much as possible out of the way of infection. These constituted his 'lot témoin,'—his standard of comparison. On April 16, 1868, he thus infected thirty worms. Up to the 23rd they remained quite well. On the 25th they seemed well, but on that day corpuscles were found in the intestines of two of them. On the 27th, or eleven days after the infected repast, two fresh worms were examined, and not only was the intestinal canal found in each case invaded, but the silk organ itself was charged with corpuscles. On the 28th the twenty-six remaining worms were covered by the black spots of pébrine. On the 30th the difference of size between the infected and non-infected worms was very striking, the sick worms being not more than two-thirds of the bulk of the healthy ones. On May 2 a worm which had just finished its fourth moulting was examined. Its whole body was so filled with the

parasite as to excite astonishment that it could live. The disease advanced, the worms died and were examined, and on May 11 only six out of the thirty remained. They were the strongest of the lot, but on being searched they also were found charged with corpuscles. Not one of the thirty worms had escaped; a single meal had poisoned them all. The standard lot, on the contrary, spun their fine cocoons, two only of their moths being proved to contain any trace of the parasite, which had doubtless been introduced during the rearing of the worms.

As his acquaintance with the subject increased, Pasteur's desire for precision augmented, and he finally counted the growing number of corpuscles seen in the field of his microscope from day to day. After a contagious repast the number of worms containing the parasite gradually augmented until finally it became cent. per cent. The number of corpuscles would at the same time rise from 0 to 1, to 10, to 100, and sometimes even to 1,000 or 1,500 in the field of his microscope. He then varied the mode of infection. He inoculated healthy worms with the corpusculous matter, and watched the consequent growth of the disease. He proved that the worms inoculate each other by the infliction of visible wounds with their claws. In various cases he washed the claws, and found corpuscles in the water. He demonstrated the spread of infection by the simple association of healthy and diseased worms. By their claws and their excrement, the diseased worms spread infection. It was no hypothetical infected medium—no problematical pythogenic gas—that killed the worms, but a definite organism. The question of infection at a distance was also examined, and its occurrence demonstrated. As might be expected from Pasteur's antecedents, the investigation was exhaustive.

the beauty of his manipulation finding a fit correlative in the clearness of his thought.

The following quotation from Pasteur's work clearly shows the relation in which his researches stand to the important question on which he was engaged :

Place (he says) the most skilful cultivator, even the most expert microscopist, in presence of large cultivations which present the symptoms described in our experiments ; his judgment will necessarily be erroneous if he confines himself to the knowledge which preceded my researches. The worms will not present to him the slightest spot of pébrine ; the microscope will not reveal the existence of corpuscles ; the mortality of the worms will be null or insignificant; and the cocoons leave nothing to be desired. Our observer would, therefore, conclude without hesitation that the eggs produced will be good for incubation. The truth is, on the contrary, that all the worms of these fine crops have been poisoned ; that from the beginning they carried in them the germ of the malady ; ready to multiply itself beyond measure in the chrysalides and the moths, thence to pass into the eggs and smite with sterility the next generation. And what is the first cause of the evil concealed under so deceitful an exterior? In our experiments we can, so to speak, touch it with our fingers. It is entirely the effect of a single corpusculous repast; an effect more or less prompt according to the epoch of life of the worm that has eaten the poisoned food.

Pasteur describes in detail his method of securing healthy eggs. It is nothing less than a mode of restoring to France her ancient silk husbandry. The justification of his work is to be found in the reports which reached him of the application and the success of his method, while editing his researches for final publication. In both France and Italy his method has been pursued with the most satisfactory results. But it was an up-hill fight which led to this victory.

'Since the commencement,' he says, 'of these researches, I have been constantly exposed to the most obstinate and unjust contradictions; but I have made it a duty to leave no trace of these conflicts in this book.' And in reference to parasitic diseases, generally, he uses the following weighty words: 'Il est au pouvoir de l'homme de faire disparaître de la surface du globe les maladies parasitaires, si, comme c'est ma conviction, la doctrine des générations spontanées est une chimère.'

Pasteur dwells upon the ease with which an island like Corsica might be absolutely isolated from the silkworm epidemic. And with regard to other epidemics, Mr. John Simon describes an extraordinary case of insular exemption, for the ten years extending from 1851 to 1860. Of the 627 registration districts of England, one only had an entire escape from diseases which, in whole or in part, were prevalent in all the others: 'In all the ten years it had not a single death by measles, nor a single death by small-pox, nor a single death by scarlet-fever. And why? Not because of its general sanitary merits, for it had an average amount of other evidence of unhealthiness. Doubtless, the reason of its escape was that it was insular. It was the district of the Scilly Isles; to which it was most improbable that any febrile contagion should come from without. And its escape is an approximative proof that, at least for those ten years, no contagium of measles, nor any contagium of scarlet-fever, nor any contagium of small-pox had arisen spontaneously within its limits.' It may be added that there were only seven districts in England in which no death from diphtheria occurred, and that, of those seven, the district of the Scilly Isles was one.

A second parasitic disease of silkworms, called in France *la flacherie*, co-existent with pébrine, but quite

distinct from it, has also been investigated by Pasteur. Enough, however, has been said to send those interested in these questions to the original volumes for further information. To one important practical point M. Pasteur, in a letter to myself, directs attention :

Permettez-moi de terminer ces quelques lignes que je dois dicter, vaincu que je suis par la maladie, en vous faisant observer que vous rendriez service aux Colonies de la Grande-Bretagne en répandant la connaissance de ce livre, et des princ'pes que j'établis touchant la maladie des vers à soie. Beaucoup de ces colonies pourraient cultiver le mûrier avec succès, et, en jetant les yeux sur mon ouvrage, vous vous convaincrez aisément qu'il est facile aujourd'hui, non-seulement d'éloigner la maladie régnante, mais en outre de donner aux récoltes de la soie une prospérité qu'elles n'ont jamais eue.

Origin and Propagation of Contagious Matter.

Prior to Pasteur, the most diverse and contradictory opinions were entertained as to the contagious character of pébrine ; some stoutly affirmed it, others as stoutly denied it. But on one point all were agreed. 'They believed in the existence of a deleterious medium, rendered epidemic by some occult and mysterious influence, to which was attributed the cause of the disease.' Those acquainted with our medical literature will not fail to observe an instructive analogy here. We have on the one side accomplished writers, like Dr. Murchison, ascribing epidemic diseases to 'deleterious media' which arise spontaneously in crowded hospitals and ill-smelling drains. According to them, the contagia of epidemic disease are formed *de novo* in a putrescent atmosphere. On the other side we have, writers like Dr. Budd, clear, vigorous, with well-defined ideas and

methods of research, contending that the matter which produces epidemic disease comes always from a parent stock. It behaves as germinal matter, and they do not hesitate to regard it as such. They no more believe in the spontaneous generation of such diseases, than they do in the spontaneous generation of mice. Pasteur, for example, found that pébrine had been known for an indefinite time as a disease among silkworms. The development of it which he combated was merely the expansion of an already existing power—the bursting into open conflagration of a previously smouldering fire. There is nothing surprising in this. For though epidemic disease requires a special contagium to produce it, surrounding conditions must have a potent influence on its development. Common seeds may be duly sown, but the conditions of temperature and moisture may be such as to restrict, or altogether prevent, the subsequent growth. Looked at, therefore, from the point of view of the germ theory, the exceptional energy which epidemic disease from time to time exhibits is in harmony with the method of Nature. We sometimes hear diphtheria spoken of as if it were a new disease ; but Mr. Simon tells me that about three centuries ago tremendous epidemics of it raged in Spain (where it was named *Garrotillo*), and soon afterwards in Italy; and that since that time the disease has been well known to all successive generations of doctors. In or about 1758, for instance, Dr. Starr, of Liskeard, in a communication to the Royal Society, particularly described the disease, with all the symptoms which have recently again become familiar to us, but under the name of *morbus strangulatorius*, as then severely epidemic in Cornwall. This fact is the more interesting, as diphtheria, in its more modern reappearance, again showed predilection for that remote county. Many also believe that the Black Death, of

five centuries ago, has disappeared as mysteriously as it came; but Mr. Simon finds that it is believed to be prevalent at this hour in some of the north-western parts of India.

Let me here state an item of my own experience. When I was at the Bel Alp in 1869, the English chaplain received letters informing him of the breaking out of scarlet-fever among his children. He lived, if I remember rightly, on the healthful eminence of Dartmoor, and it was difficult to imagine how scarlet-fever could have been wafted to the place. A drain ran close to his house, and on it his suspicions were manifestly fixed. Some of our medical writers would fortify him in this notion, and thus deflect him from the truth, while those of another, and, in my opinion, a wiser school, would deny to a drain, however foul, the power of generating *de novo* a specific disease. After close inquiry he recollected that a hobby-horse had been used both by his boy and by another who, a short time previously, had passed through scarlet-fever.

Drains and cesspools, indeed, are by no means in such evil odour as they used to be. A fetid Thames and a low death-rate occur from time to time together in London. For, if the special matter or germs of epidemic disorder be not present, a corrupt atmosphere, however obnoxious otherwise, will not produce the disorder. But, if the germs be present, defective drains and cesspools become the potent distributors of disease and death. Corrupted air may promote an epidemic, but cannot produce it. On the other hand, through the transport of the special germ or virus, disease may develop itself in regions where the drainage is good and the atmosphere pure.

If you see a new thistle growing in your field, you feel sure that its seed has been wafted thither. Just as

sure does it seem that the contagious matter of epidemic disease has been sown in the place where it newly appears. With a clearness and conclusiveness not to be surpassed, Dr. William Budd has traced such diseases from place to place; showing how they plant themselves, at distinct points, among populations subjected to the same atmospheric influences, just as grains of corn might be carried in the pocket and sown. Hildebrand, to whose remarkable work, 'Du Typhus contagieux,' Dr. de Mussy has directed my attention, gives the following striking case, both of the transport and the durability of the virus of scarlatina: 'Un habit noir que j'avais en visitant une malade attaquée de scarlatine, et que je portai de Vienne en Podolie, sans l'avoir mis depuis plus d'un an et demi, me communiqua, dès que je fus arrivé, cette maladie contagieuse, que je répandis ensuite dans cette province, où elle était jusqu'alors presque inconnue.' Some years ago Dr. de Mussy himself was summoned to a country house in Surrey, to see a young lady who was suffering from a dropsy, evidently the consequence of scarlatina. The original disease, being of a very mild character, had been quite overlooked; but circumstances were recorded which could leave no doubt upon the mind as to the nature and cause of the complaint. But then the question arose, How did the young lady catch the scarlatina? She had come there on a visit two months previously, and it was only after she had been a month in the house that she was taken ill. The housekeeper at length cleared up the mystery. The young lady, on her arrival, had expressed a wish to occupy a room in an isolated tower. Her desire was granted; and in that room, six months previously, a visitor had been confined with an attack of scarlatina. The room had been swept and whitewashed, but the carpets had been permitted to remain.

Thousands of cases could probably be cited in which the disease has shown itself in this mysterious way, but where a strict examination has revealed its true parentage and extraction. Is it, then, philosophical to take refuge in the fortuitous concourse of atoms as a cause of specific disease, merely because in special cases the parentage may be indistinct? Those best acquainted with atomic nature, and who are most ready to admit, as regards even higher things than this, the potentialities of matter, will be the last to accept these rash hypotheses.

The Germ Theory applied to Surgery.

Not only medical but still more especially surgical science is now seeking light and guidance from this germ theory. Upon it the antiseptic system of Professor Lister of Edinburgh is founded. As already stated, the germ theory of putrefaction was started by Schwann; but the illustrations of this theory adduced by Professor Lister are of such public moment as not only to justify, but to render imperative, their introduction here.

Schwann's observations (says Professor Lister) did not receive the attention which they appeared to me to have deserved. The fermentation of sugar was generally allowed to be occasioned by the *torùla cerevisiæ*; but it was not admitted that putrefaction was due to an analogous agency. And yet the two cases present a very striking parallel. In each a stable chemical compound, sugar in the one case, albumen in the other, undergoes extraordinary chemical changes under the influence of an excessively minute quantity of a substance which, regarded chemically, we should suppose inert. As an example of this in the case of putrefaction, let us take a circumstance often witnessed in the treatment

of large chronic abscesses. In order to guard against the access of atmospheric air, we used to draw off the matter by means of a canula and trocar, such as you see here, consisting of a silver tube with a sharp-pointed steel rod fitted into it, and projecting beyond it. The instrument, dipped in oil, was thrust into the cavity of the abscess, the trocar was withdrawn, and the pus flowed out through the canula, care being taken by gentle pressure over the part to prevent the possibility of regurgitation. The canula was then drawn out with due precaution against the reflux of air. This method was frequently successful as to its immediate object, the patient being relieved from the mass of the accumulated fluid, and experiencing no inconvenience from the operation. But the pus was pretty certain to reaccumulate in course of time, and it became necessary again and again to repeat the process. And unhappily there was no absolute security of immunity from bad consequences. However carefully the procedure was conducted, it sometimes happened, even though the puncture seemed healing by first intention, that feverish symptoms declared themselves in the course of the first or second day, and, on inspecting the seat of the abscess, the skin was perhaps seen to be red, implying the presence of some cause of irritation, while a rapid reaccumulation of the fluid was found to have occurred. Under these circumstances, it became necessary to open the abscess by free incision, when a quantity, large in proportion to the size of the abscess, say, for example, a quart, of pus escaped, fetid from putrefaction. Now, how had this change been brought about? Without the germ theory, I venture to say, no rational explanation of it could have been given. It must have been caused by the introduction of something from without. Inflammation of the punctured wound, even supposing it to have occurred, would not explain the phenomenon. For mere inflammation, whether acute or chronic, though it occasions the formation of pus, does not induce putrefaction. The pus originally evacuated was perfectly sweet, and we know nothing to account for the alteration in its quality but the influence of something derived from

the external world. And what could that something be? The dipping of the instrument in oil, and the subsequent precautions, prevented the entrance of oxygen. Or even if you allowed that a few atoms of the gas did enter, it would be an extraordinary assumption to make that these could in so short a time effect such changes in so large a mass of albuminous material. Besides, the pyogenic membrane is abundantly supplied with capillary vessels, through which arterial blood, rich in oxygen, is perpetually flowing; and there can be little doubt that the pus, before it was evacuated at all, was liable to any action which the element might be disposed to exert upon it.

On the oxygen theory, then, the occurrence of putrefaction under these circumstances is quite inexplicable. But if you admit the germ theory, the difficulty vanishes at once. The canula and trocar having been lying exposed to the air, dust will have been deposited upon them, and will be present in the angle between the trocar and the silver tube, and in that protected situation will fail to be wiped off when the instrument is thrust through the tissues. Then when the trocar is withdrawn, some portions of this dust will naturally remain upon the margin of the canula, which is left projecting into the abscess, and nothing is more likely than that some particles may fail to be washed off by the stream of out-flowing pus, but may be dislodged when the tube is taken out, and left behind in the cavity. The germ theory tells us that these particles of dust will be pretty sure to contain the germs of putrefactive organisms, and if one such is left in the albuminous liquid, it will rapidly develop at the high temperature of the body, and account for all the phenomena.

But striking as is the parallel between putrefaction in this instance and the vinous fermentation, as regards the greatness of the effect produced, compared with the minuteness and the inertness, chemically speaking, of the cause, you will naturally desire further evidence of the similarity of the two processes. You can see with the microscope the torula of fermenting must or beer. Is there, you may ask, any

organism to be detected in the putrefying pus? Yes, gentlemen, there is. If any drop of the putrid matter is examined with a good glass, it is found to be teeming with myriads of minute jointed bodies, called vibrios, which indubitably proclaim their vitality by the energy of their movements. It is not an affair of probability, but a fact, that the entire mass of that quart of pus has become peopled with living organisms as the result of the introduction of the canula and trocar; for the matter first let out was as free from vibrios as it was from putrefaction. If this be so, the greatness of the chemical changes that have taken place in the pus ceases to be surprising. We know that it is one of the chief peculiarities of living structures that they possess extraordinary powers of effecting chemical changes in materials in their vicinity, out of all proportion to their energy as mere chemical compounds. And we can hardly doubt that the animalcules which have been developed in the albuminous liquid, and have grown at its expense, must have altered its constitution, just as we ourselves alter that of the materials on which we feed.[1]

In antiseptic operations care is taken that every portion of tissue laid bare by the knife shall be defended from germs; that if they fall upon the wound they should be killed as they fall. With this in view he showers upon his exposed surfaces the spray of dilute carbolic acid, which is particularly deadly to the germs, and he surrounds the wound in the most careful manner with antiseptic bandages. To those accustomed to strict experiment it is manifest that we have a strict experimenter here—a man with a perfectly distinct object in view, which he pursues with never-tiring patience and unwavering faith. And the result, in his hospital practice, as described by himself, has been, that even in the midst of abominations too shocking to be mentioned here, and in the neighbourhood of

[1] Introductory Lecture before the University of Edinburgh.

wards where death was rampant from pyæmia, erysipelas, and hospital gangrene, he was able to keep his patients absolutely free from these terrible scourges. Let me here recommend to your attention Professor Lister's 'Introductory Lecture before the University of Edinburgh,' which I have already quoted; his paper on 'The Effect of the Antiseptic System of Treatment on the Salubrity of a Surgical Hospital;' and the article in the 'British Medical Journal' of January 14, 1871.

If, instead of using carbolic acid spray, he could surround his wounds with properly filtered air, the result would, he contends, be the same. In a room, where the germs not only float but cling to clothes and walls, this would be difficult, if not impossible. But surgery is acquainted with a class of wounds in which the blood is freely mixed with air that has passed through the lungs, and it is a most remarkable fact that such air does not produce putrefaction. Professor Lister, as far as I know, was the first to give a philosophical interpretation of this fact, which he describes and comments upon thus:

I have explained to my own mind the remarkable fact that in simple fracture of the ribs, if the lung be punctured by a fragment, the blood effused into the pleural cavity, though freely mixed with air, undergoes no decomposition. The air is sometimes pumped into the pleural cavity in such abundance that, making its way through the wound in the pleura costalis, it inflates the cellular tissue of the whole body. Yet this occasions no alarm to the surgeon (although if the blood in the pleura were to putrefy, it would infallibly occasion dangerous suppurative pleurisy). Why air introduced into the pleural cavity through a wounded lung, should have such wholly different effects from that entering directly through a wound in the chest, was to me a complete mystery until I heard of the germ theory of putrefaction, when it at once occurred to me that it was only natural that air should

be filtered of germs by the air-passages, one of whose offices is to arrest inhaled particles of dust, and prevent them from entering the air-cells.

I shall have occasion to refer to this remarkable hypothesis farther on.

The advocates of the germ theory, both of putrefaction and epidemic disease, hold that both arise, not from the air, but from something contained in the air. They hold, moreover, that this 'something' is not a vapour nor a gas, nor indeed a molecule of any kind, but a *particle*.[1] The term 'particulate' has been used by Mr. Simon in the Reports of the Medical Department of the Privy Council to describe this supposed constitution of contagious matter; and Dr. Sanderson's experiments render it in the highest degree probable, if they do not actually demonstrate, that the virus of small-pox is 'particulate.' Definite knowledge upon this point is of exceeding importance, because in the treatment of *particles* methods are available which it would be futile to apply to *molecules*.

The Luminous Beam as a means of Research.

My own interference with this great question, while sanctioned by eminent names, has been also an object of varied and ingenious attack. On this point I will only say that when angry feeling escapes from behind the intellect, where it may be useful as an urging force, and places itself athwart the intellect, it is liable to

[1] As regards size, there is probably no sharp line of division between molecules and particles; the one gradually shades into the other. But the distinction that I draw is this: the atom or the molecule, if free, is always part of a gas, the particle is never so. A particle is a bit of liquid or solid matter, formed by the aggregation of atoms or molecules.

produce all manner of delusions. Thus my censors, for the most part, have levelled their remarks against positions which were never assumed, and against claims which were never made. The simple history of the matter is this: During the autumn of 1868 I was much occupied with the observations referred to at the beginning of this discourse, and in part described in the preceding article. For fifteen years it had been my habit to make use of floating dust to reveal the paths of luminous beams through the air; but until 1868 I did not intentionally reverse the process, and employ a luminous beam to reveal and examine the dust. In a paper presented to the Royal Society in December, 1869, the observations which induced me to give more special attention to the question of spontaneous generation, and the germ theory of epidemic disease, are thus described:

The Floating Matter of the Air.

Prior to the discovery of the foregoing action (the chemical action of light upon vapours), and also during the experiments just referred to, the nature of my work compelled me to aim at obtaining experimental tubes absolutely clean upon the interior surface, and absolutely free within from suspended matter. Neither condition is, however, easily attained.

For however well the tubes might be washed and polished, and however bright and pure they might appear in ordinary daylight, the electric beam infallibly revealed signs and tokens of dirt. The air was always present, and it was sure to deposit some impurity. All chemical processes, not conducted in a vacuum, are open to this disturbance. When the experimental tube was exhausted, it exhibited no trace of floating matter, but on admitting the air through two U-tubes containing respectively caustic potash and sulphuric acid, a

dust-cone more or less distinct was always revealed by the powerfully condensed electric beam.

The floating motes resembled minute particles of liquid which had been carried mechanically from the U-tubes into the experimental tube. Precautions were therefore taken to prevent any such transfer. They produced little or no mitigation. I did not imagine, at the time, that the dust of the external air could find such free passage through the caustic potash and sulphuric acid. This, however, was the case; the motes really came from without. They also passed with freedom through a variety of æthers and alcohols. In fact, it requires long-continued action on the part of an acid first to *wet* the motes and afterwards to destroy them. By carefully passing the air through the flame of a spirit lamp, or through a platinum tube heated to bright redness, the floating matter was sensibly destroyed. It was therefore combustible, in other words, *organic*, matter. I tried to intercept it by a large respirator of cotton-wool. Close pressure was necessary to render the wool effective. A plug of the wool, rammed pretty tightly into the tube through which the air passed, was finally found competent to hold back the motes. They appeared from time to time afterwards, and gave me much trouble; but they were invariably traced in the end to some defect in the purifying apparatus —to some crack or flaw in the sealing-wax employed to render the tubes air-tight. Thus through proper care, but not without a great deal of searching out of disturbances, the experimental tube, even when filled with air or vapour, contains nothing competent to scatter the light. The space within it has the aspect of an absolute vacuum.

An experimental tube in this condition I call *optically empty*.

The facts here forced upon my attention had a bearing too evident to be overlooked. The inability of air which had been filtered through cotton-wool to generate microscopic life, had been demonstrated by Schroeder and Pasteur: here the cause of its impotence was ren-

dered evident to the eye. The experiment proved that no sensible amount of light was scattered by the *molecules* of the air; that the scattered light always arose from suspended *particles*. The fact moreover that the removal of these abolished simultaneously the power of scattering light and of originating life, obviously detached the life-originating power from the air, and fixed it on something suspended in the air. Gases of all kinds passed with freedom through the plug of cotton-wool; hence the thing whose removal by the cotton-wool rendered the gas impotent, could not itself have been matter in the gaseous condition. It at once occurred to me that the retina, protected as it was, in these experiments, from all extraneous light, might be converted into a new and powerful instrument of demonstration in relation to the germ theory.

The observations just described also revealed the danger incurred in experiments of this nature; showing that without an amount of care far beyond that hitherto bestowed upon them, such experiments left the door open to errors of the gravest description. It was especially manifest that the chemical method employed by Schultze in his experiments, and so often resorted to since, might lead to the most erroneous consequences; that neither acids nor alkalies had the power of rapid destruction hitherto ascribed to them. In short, the employment of the luminous beam rendered evident the cause of success in experiments rigidly conducted like those of Pasteur; while it made equally evident the certainty of failure in experiments less severely carried out.

Dr. Bennett's Experiments.

I do not wish to leave an assertion of this kind without proof or illustration. Take, then, the well-conceived experiments of Dr. Hughes Bennett, described before the Royal Society of Surgeons in Edinburgh on January 17, 1868.[1] Into flasks containing decoctions of liquorice-root, hay, or tea, Mr. Bennett, by an ingenious method, forced air. The air was driven through two U-tubes, the one containing a solution of caustic potash, the other sulphuric acid. 'All the bent tubes were filled with fragments of pumice-stone to break up the air, so as to prevent the possibility of any germs passing through in the centre of bubbles.' The air also passed through a Liebig's bulb containing sulphuric acid, and also through a bulb containing gun-cotton.

It was only natural for Dr. Bennett to believe that his 'bent tubes' entirely cut off the germs. Previous to the observations just referred to, I also believed in their efficacy. But these observations destroy any such notion. The gun-cotton, moreover, will fail to arrest the whole of the floating matter, unless it is tightly packed, and there is no indication in Dr. Bennett's memoir that it was so packed. On the whole, I should infer, from the mere inspection of Dr. Bennett's apparatus, the very results which he has described—a retardation of the development of life, a total absence of it in some cases, and its presence in others.

In his first series of experiments, eight flasks were fed with sifted air, and five with common air. In ten or twelve days all the five had fungi in them; whilst it required from four to nine months to develop fungi in the others. In one of the eight, moreover,

[1] British Medical Journal, 13, pt. ii. 1868.

even after this interval no fungi appeared. In a second series of experiments there was a similar exception. In a third series the cork stoppers used in the first and second series were abandoned, and glass stoppers employed. Flasks containing decoctions of tea, beef, and hay were filled with common air, and other flasks with sifted air. In every one of the former fungi appeared and in not one of the latter. These experiments simply ruin the doctrine that Dr. Bennett finally espouses.

In all these negative cases, the air was forced through the bent tubes and bulb into the boiling-hot infusion. Dr. Bennett made a fourth series of experiments, in which, previous to forcing in the air, he permitted the flasks to cool. Into four bottles thus treated he forced prepared air, and after a time found fungi in all of them. What is his conclusion? Not that the boiling-hot liquid, employed in his first experiments, had destroyed such germs as had run the gauntlet of his apparatus; but that air, which, previous to being sealed up, had been exposed to a temperature of 212°, *is too rare to support life.* This conclusion is so remarkable that it ought to be stated in Dr. Bennett's own words. 'It may be easily conceived that air subjected to a boiling temperature is so expanded as scarcely to merit the name of air, and that it is more or less unfit for the purpose of sustaining animal or vegetable life.'

Numerical data are attainable here, but they are unnecessary. As a matter of fact, I live and flourish for a considerable portion of each year in a medium of less density than that which Dr. Bennett describes as scarcely meriting the name of air. The inhabitants of the higher Alpine châlets, with their flocks and herds, and the grasses which support these, do the same; while the chamois rears its kids in air rarer still.

Insect life, moreover, is sometimes exhibited with monstrous prodigality at Alpine heights.

In a fifth series of experiments sixteen bottles were filled with infusions. Into four of them, while cold, ordinary unheated and unsifted air was pumped, and in these fungi were developed. Into four other bottles, containing a boiling infusion, ordinary air was also pumped—no fungi were here developed. Into four other bottles containing an infusion which had been boiled and permitted to cool, sifted air was pumped —no fungi were developed. Finally, into four bottles containing a boiling infusion sifted air was pumped— no fungi were developed. Only, therefore, in the four cases where the infusions were cold infusions, and the air ordinary air, did fungi appear.

Dr. Bennett does not draw from his experiments the conclusion to which they so obviously point. On the contrary, he founds on them a defence of the doctrine of spontaneous generation, and a general theory of spontaneous development. So strongly was he impressed with the idea that the germs could not possibly pass through his potash and sulphuric acid tubes, that the appearance of fungi, even in a small minority of cases, where the air had been sent through these tubes, was to him conclusive evidence of the spontaneous origin of such fungi. And he accounts for the absence of life in many of his experiments by an explanation which will not bear a moment's criticism. But knowing, as we now do, that organic particles may pass unscathed through alkalies and acids, the results of Dr. Bennett are precisely what ought under the circumstances to be expected. Indeed, their harmony with the conditions now revealed is a proof of the honesty and accuracy with which they were executed.

The caution exercised by Pasteur both in the exe-

cution of his experiments, and in the reasoning based upon them, is perfectly evident to those who, through the practice of severe experimental inquiry, have rendered themselves competent to judge of good experimental work. He found germs in the mercury used to isolate his air. He was never sure that they did not cling to the instruments he employed, or to his own person. Thus when he opened his hermetically-sealed flasks upon the Mer de Glace, he had his eye upon the file used to detach the drawn-out necks of his bottles; and he was careful to stand to leeward when each flask was opened. Using these precautions, he found the glacier air incompetent, in nineteen cases out of twenty, to generate life; while similar flasks, opened amid the vegetation of the lowlands, were soon crowded with living things. M. Pouchet repeated Pasteur's experiments in the Pyrenees, adopting the precaution of holding his flasks above his head, and obtaining a different result. Now great care would be needed to render this procedure a real precaution. The luminous beam at once shows us its possible effect. Let smoking brown paper be placed at the open mouth of a glass shade, so that the smoke shall ascend and fill the shade. A beam sent through the shade forms a bright track through the smoke. When the closed fist is placed underneath the shade, a vertical wind of surprising violence, considering the small elevation of temperature, rises from the hand, displacing by comparatively dark air the illuminated smoke. Unless special care were taken such a wind would rise from M. Pouchet's body as he held his flasks above his head, and thus the precaution of Pasteur, of not coming between the wind and the flask, would be annulled.

Let me now direct attention to another result of Pasteur, the cause and significance of which are at once

revealed by the luminous beam. He prepared twenty-one flasks, each containing a decoction of yeast, filtered and clear. He boiled the decoction so as to destroy whatever germs it might contain, and while the space above the liquid was filled with pure steam, he sealed his flasks with a blow-pipe. He opened ten of them in the deep, damp caves of the Paris Observatory, and eleven of them in the courtyard of the establishment. Of the former, one only showed signs of life subsequently. In nine out of the ten flasks no organisms of any kind were developed. In all the others organisms speedily appeared.

Now here is an experiment conducted in Paris, on which we can throw light in London. Causing our luminous beam to pass through a large flask filled with the air of this room, and charged with its germs and its dust, the beam is seen crossing the flask from side to side. But here is another similar flask, which cuts a clear gap out of the beam. It is filled with *unfiltered* air, and still no trace of the beam is visible. Why? By pure accident I stumbled on this flask in our apparatus room, where it had remained quiet for some time. Acting upon this obvious suggestion, I set aside three other flasks, filled, in the first instance, with mote-laden air. They are now optically empty. Our former experiments proved that the life-producing particles attach themselves to the fibres of cotton-wool. In the present experiment the motes have been brought by gentle air-currents, established by slight differences of temperature within our closed vessels, into contact with the interior surface, to which they adhere. The air of these flasks has deposited its dust, germs and all, and is practically free from suspended matter.

I had a chamber erected, the lower half of which is of wood, its upper half being enclosed by four glazed

window-frames. It tapers to a truncated cone at the top. It measures in plan 3 ft. by 2 ft. 6 in., and its height is 5 ft. 10 in. On February 6 it was closed, every crevice that could admit dust, or cause displacement of the air, being carefully pasted over with paper. The electric beam at first revealed the dust within the chamber as it did in the air of the laboratory. The chamber was examined almost daily; a perceptible diminution of the floating matter being noticed as time advanced. At the end of a week the chamber was optically empty, exhibiting no trace of matter competent to scatter the light. Such must have been the case in the stagnant caves of the Paris Observatory. Were our electric beam sent through the air of these caves, its track would, doubtless, save from aqueous haze, be invisible; thus showing the indissoluble association of the scattering of light by air and its power to generate life.

I will now turn to what seems to me a more interesting application of the luminous beam than any hitherto described. My reference to Professor Lister's interpretation of the fact, that air which has passed through the lungs cannot produce putrefaction, is fresh in your memories. 'Why air,' said he, 'introduced into the pleural cavity, through a wounded lung, should have such wholly different effects from that entering through a permanently open wound, penetrating from without, was to me a complete mystery, till I heard of the germ theory of putrefaction, when it at once occurred to me that it was only natural that the air should be filtered of germs by the air passages, one of whose offices is to arrest inhaled particles of dust, and prevent them from entering the air-cells.'

Here is a surmise which bears the stamp of genius but which needs verification. If, for the words 'it is only natural' we were authorized to write 'it is per-

fectly certain,' the demonstration would be complete. Such demonstration is furnished by experiments with a beam of light. One evening, towards the close of 1869, while pouring various pure gases across the dusty track of a luminous beam, the thought occurred to me of using my breath instead of the gases. I then noticed, for the first time, the extraordinary darkness produced by the expired air, *towards the end of the expiration.* Permit me to repeat the experiment in your presence. I fill my lungs with ordinary air and breathe through a glass tube across the beam. The condensation of the aqueous vapour of the breath is shown by the formation of a luminous white cloud of delicate texture. We abolish this cloud by drying the breath previous to its entering the beam; or, still more simply, by warming the glass tube. The luminous track of the beam is for a time uninterrupted by the breath, because the dust returning from the lungs makes good, in great part, the particles displaced. After a time, however, an obscure disk appears in the beam, the darkness of which increases, until finally, towards the end of the expiration, the beam is, as it were, pierced by an intensely black hole, in which no particles whatever can be discerned. The deeper air of the lungs is thus proved to be absolutely free from suspended matter. It is therefore in the precise condition required by Professor Lister's explanation. This experiment may be repeated any number of times with the same result. I think it must be regarded as a crowning piece of evidence both of the correctness of Professor Lister's views and of the impotence, as regards vital development, of optically pure air.[1]

[1] Dr. Burdon Sanderson draws attention to the important observation of Brauell, which shows that the contagium of a pregnant animal, suffering from splenic fever, is not found in the blood of the fœtus; the placental apparatus acting as a filter, and holding back the infective particles.

The foregoing essay, as far as it relates to the theory which ascribes epidemic disease to the development of low parasitic life within the human life, was embodied in a discourse delivered before the Royal Institution in January 1870. In June 1871, after a brief reference to the polarization of light by cloudy matter, I ventured to recur to the subject in these terms: What is the practical use of these curiosities? If we exclude the interest attached to the observation of new facts, and the enhancement of that interest through the knowledge that facts often become the exponents of laws, these curiosities are in themselves worth little. They will not enable us to add to our stock of food, or drink, or clothes, or jewellery. But though thus shorn of all usefulness in themselves, they may, by carrying thought into places which it would not otherwise have entered, become the antecedents of practical consequences. In looking, for example, at our illuminated dust, we may ask ourselves what it is. How does it act, not upon a beam of light, but upon our own bodies? The question then assumes a practical character. We find on examination that this dust is mainly organic matter—in part living, in part dead. There are among it particles of ground straw, torn rags, smoke, the pollen of flowers, the spores of fungi, and the germs of other things. But what have they to do with the animal economy? Let me give you an illustration to which my attention has been lately drawn by Mr. George Henry Lewes, who writes to me thus:

'I wish to direct your attention to the experiments of Von Recklingshausen, should you happen not to know them. They are striking confirmations of what you say of dust and disease. Last spring, when I was at his laboratory in Würzburg, I examined with him blood that had been three weeks, a month, and five weeks, out

of the body, preserved in little porcelain cups under glass shades. This blood was living and growing. Not only were the Amœba-like movements of the white corpuscles present, but there were abundant evidences of the growth and development of the corpuscles. I also saw a frog's heart still pulsating which had been removed from the body (I forget how many days, but certainly more than a week). There were other examples of the same persistent vitality, or absence of putrefaction. Von Recklingshausen did not attribute this to the absence of germs—germs were not mentioned by him; but when I asked him how he represented the thing to himself, he said the whole mystery of his operation consisted in keeping the blood *free from dirt*. The instruments employed were raised to a red heat just before use; the thread was silver thread and was similarly treated; and the porcelain cups, though not kept free from air, were kept free from currents. He said he often had failures, and these he attributed to particles of dust having escaped his precautions.'

Professor Lister, who has founded upon the removal or destruction of this 'dirt' momentous improvements in surgery, tells us the effect of its introduction into the blood of wounds. The blood putrefies and becomes fetid; and when you examine more closely what putrefaction means, you find the putrefying substance swarming with infusorial life, the germs of which have been derived from the atmospheric dust.

We are now assuredly in the midst of practical matters; and with your permission I will refer once more to a question which has recently occupied a good deal of public attention. As regards the lowest forms of life, the world is divided, and has for a long time been divided, into two parties, the one affirming that we have only to submit absolutely dead matter to certain

physical conditions, to evolve from it living things; the other (without wishing to set bounds to the power of matter) affirming that, *in our day*, life has never been found to arise independently of pre-existing life. I belong to the party which claims life as a derivative of life. The question has two factors—the evidence, and the mind that judges of the evidence; and it may be purely a mental set or bias on my part that causes me, throughout this long discussion, to see, on the one side, dubious facts and defective logic, and on the other side firm reasoning and a knowledge of what rigid experimental inquiry demands. But, judged of practically, what, again, has the question of Spontaneous Generation to do with us? Let us see. There are numerous diseases of men and animals that are demonstrably the products of parasitic life, and such diseases may take the most terrible epidemic forms, as in the case of the silkworms of France, referred to at an earlier part of this essay. Now it is in the highest degree important to know whether the parasites in question are spontaneously developed, or whether they have been wafted from without to those afflicted with the disease. The means of prevention, if not of cure, would be widely different in the two cases.

But this is not all. Besides these universally admitted cases, there is the broad theory, now broached and daily growing in strength and clearness—daily, indeed, gaining more and more of assent from the most successful workers and profound thinkers of the medical profession itself—the theory, namely, that contagious disease, generally, is of this parasitic character. Had I any cause to regret having introduced this theory to your notice more than a year ago, that regret should now be expressed. I would certainly renounce in your presence whatever leaning towards the germ theory

my words might then have betrayed. But since the time referred to nothing has occurred to shake my conviction of the truth of the theory. Let me briefly state the grounds on which its supporters rely. From their respective viruses you may plant typhoid fever, scarlatina, or small-pox. What is the crop that arises from this husbandry? As surely as a thistle rises from a thistle seed, as surely as the fig comes from the fig, the grape from the grape, the thorn from the thorn, so surely does the typhoid virus increase and multiply into typhoid fever, the scarlatina virus into scarlatina, the small-pox virus into small-pox. What is the conclusion that suggests itself here? It is this: That the thing which we vaguely call a virus is to all intents and purposes a *seed*. Excluding the notion of vitality, in the whole range of chemical science you cannot point to an action which illustrates this perfect parallelism with the phenomena of life—this demonstrated power of self-multiplication and reproduction. The germ theory alone accounts for the phenomena.

In cases of epidemic disease, it is not on bad air or foul drains that the attention of the physician of the future will primarily be fixed, but upon disease germs, which no bad air or foul drains can create, but which may be pushed by foul air into virulent energy of reproduction. You may think I am treading on dangerous ground, that I am putting forth views that may interfere with salutary practice. No such thing. If you wish to learn the impotence of medical practice in dealing with contagious diseases, you have only to refer to the Harveian oration for 1871, by Sir William Gull. Such diseases defy the physician. They must run their course, and the utmost that can be done for them is careful nursing. And this, though I do not specially insist

upon it, would favour the idea of their vital origin. For if the seeds of contagious disease be themselves living things, it may be difficult to destroy either them or their progeny, without involving their living habitat in the same destruction.

It has been said, and it is sure to be repeated, that I am quitting my own *métier*, in speaking of these things. Not so. I am dealing with a question on which minds accustomed to weigh the value of experimental evidence are alone competent to decide, and regarding which, in its present condition, minds so trained are as capable of forming an opinion as regarding the phenomena of magnetism or radiant heat. 'The germ theory of disease,' it has been said, 'appertains to the biologist and the physician.' Where, I would ask in reply, is the biologist or physician, whose researches, in connection with this subject, could for one instant be compared to those of the chemist Pasteur? It is not the philosophic members of the medical profession who are dull to the reception of truth not originated within the pale of the profession itself. I cannot better conclude this portion of my story than by reading to you an extract from a letter addressed to me some time ago by Dr. William Budd, of Clifton, to whose insight and energy the town of Bristol owes so much in the way of sanitary improvement.

'As to the germ theory itself,' writes Dr. Budd, 'that is a matter on which I have long since made up my mind. From the day when I first began to think of these subjects I have never had a doubt that the specific cause of contagious fevers must be living organisms.

'It is impossible, in fact, to make any statement bearing upon the essence or distinctive characters of these fevers, without using terms which are of all others *the most distinctive of life*. Take up the writings of

the most violent opponent of the germ theory, and, ten to one, you will find them full of such terms as "propagation," "self-propagation," "reproduction," "self-multiplication," and so on. Try as he may—if he has anything to say of those diseases which is characteristic of them—he cannot evade the use of these terms, or the exact equivalents to them. While perfectly applicable to living things, these terms express qualities which are not only inapplicable to common chemical agents, but, as far as I can see, actually inconceivable of them.'

OPTICAL DEPORTMENT OF THE ATMOSPHERE

IN RELATION TO

PUTREFACTION AND INFECTION.[1]

II.

§ 1. *Introduction.*

An inquiry into the decomposition of vapours by light, begun in 1868 and continued in 1869,[2] in which it was necessary to employ optically pure air, led me to experiment on the floating matter of the atmosphere. A brief section of a paper published in the Philosophical Transactions for 1870[3] is devoted to this subject.

I at that time found that the air of London rooms, which is always thick with motes, and also with matter too fine to be described as motes, after it had been filtered by passing it through densely packed cotton-wool, or calcined by passing it through a red-hot platinum-tube containing a bundle of red-hot platinum wires, or by carefully leading it over the top of a spirit-lamp flame, showed, when examined by a concentrated luminous beam, no trace of mechanically suspended matter. The particular portion of space occupied by such a beam was not to be distinguished from adjacent space.

The purely gaseous portion of our atmosphere was thus shown to be incompetent to scatter light.

[1] Philosophical Transactions, Part I., 1876.

[2] Proc. Roy. Soc. vol. xvii. [3] Vol. clx. p. 337.

I subsequently found that, to render the air thus optically pure, it was only necessary to leave it to itself for a sufficient time in a small closed chamber, or in a suitably closed vessel. The floating matter gradually attached itself to the top and sides, or sank to the bottom, leaving behind it air possessing no scattering power. Sent through such air, the most concentrated beam failed to render its track visible.

I mention 'top' and 'sides,' as well as 'bottom,' because gravity is not the only agent, perhaps not even the principal agent, concerned in the removal of the floating matter. It is practically impossible to surround a closed vessel by an absolutely uniform temperature; and where differences of temperature, however small, exist, air-currents will be established. By such gentle currents the floating particles are gradually brought into contact with all the surrounding surfaces. To these they adhere, and, no new supply being admitted, the suspended matter finally disappears from the air altogether.

The striking parallelism of these results with those obtained in the excellent researches of Schwann,[1] Schroeder and Dusch,[2] Schroeder himself,[3] and Pasteur[4] in regard to the question of 'spontaneous generation,' caused me to conclude that the power of scattering light, and the power of producing life, by atmospheric air would be found to go hand in hand.

This conclusion was strengthened by an experiment easily made and of high significance in relation to this question. It had been pointed out by Professor Lister[5]

[1] Pogg. Ann. 1837, vol. xli. p. 184.

[2] Ann. der Pharmacie, vol. lxxxix. p. 232.

[3] *Ibid.* vol. cix. p. 35.

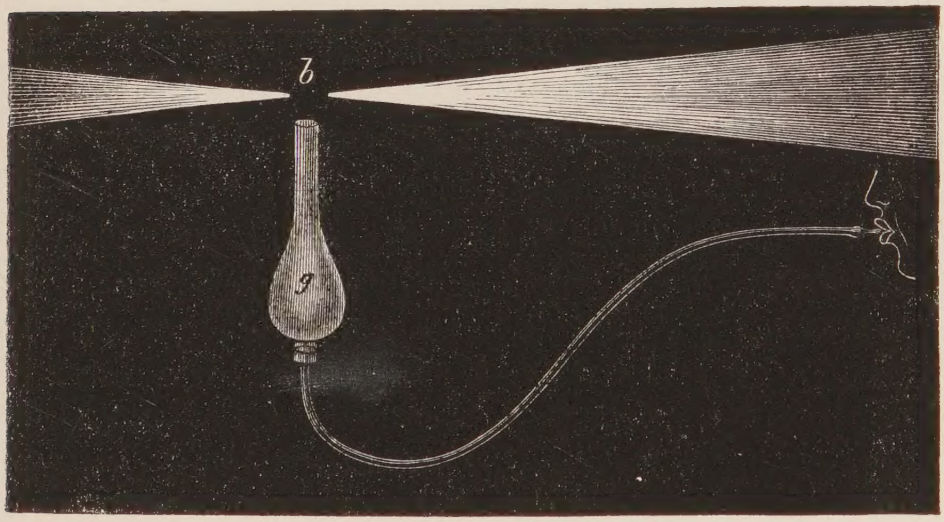
[4] Ann. de Chim. et de Phys. 3rd series, vol. lxiv. p. 83.

[5] Introductory Lecture before the University of Edinburgh.

that air which has passed through the lungs is known to have lost its power of causing putrefaction. Such air may mix freely with the blood of an internal wound without risk of mischief; and that truly great scientific surgeon had the penetration to ascribe this immunity from danger to the filtering power of the lungs. Prior to my becoming acquainted with this hypothesis in 1869, I had demonstrated its accuracy in the following manner.[1]

Condensing in a dark room, and in dusty air, a

Fig. 1.



powerful beam of light, and breathing through a glass tube (the tube actually employed was a lamp-glass, rendered warm in a flame to prevent condensation of the breath) across the focus, a diminution of the scattered light was first observed. But towards the end of the expiration the white track of the beam was broken by a perfectly black gap, the blackness being due to the total absence from the expired air of any matter competent to scatter light. The experimental arrangement is represented in fig. 1, where *g* represents the heated

[1] Proc. Roy. Inst. vol. vi. p. 9.

lamp-glass, and *b* the black gap cut out of the beam at its brightest point. The deeper portions of the lungs were thus proved to be filled with optically pure air, which, as such, had no power to generate the organisms proved by Schwann to be essential to the process of putrefaction.[1]

It seemed that this simple method of examination could not fail to be of use to workers in this field. They had hitherto proceeded less by sight than by insight, being in general unable to observe the physical character of the medium in which their experiments were conducted. But the method has not been much turned to account; and this year (1875) I thought it worth while to devote some time myself to the more complete demonstration of its utility.

I also wished to free my mind, and if possible the minds of others, from the uncertainty and confusion which now beset the doctrine of 'spontaneous generation.' Pasteur has pronounced it 'a chimera,' and expressed the undoubting conviction that this being so it is possible to remove all parasitic diseases from the earth. To the medical profession, therefore, and through them to humanity at large, this question, if the illustrious French philosopher be correct, is one of the last importance. But Pasteur's labours, which have so long been considered models by most of us, have been subjected to

[1] 'No putrefaction,' says Cohn, 'can occur in a nitrogenous substance if it be kept free from the entrance of new *Bacteria* after those which it may contain have been destroyed. Putrefaction begins as soon as *Bacteria*, even in the smallest numbers, are accidentally or purposely introduced. It progresses in direct proportion to the multiplication of the *Bacteria*; it is retarded when the *Bacteria* (for example, by a low temperature) develop a small amount of vitality, and is brought to an end by all influences which either stop the development of the *Bacteria*, or kill them. All bactericidal media are therefore antiseptic and disinfecting.'—*Beiträge zur Biologie der Pflanzen*, zweites Heft, 1872, p. 203.

rough handling of late. His reasoning has been criticised, and experiments counter to his have been adduced in such number and variety, and with such an appearance of circumstantial accuracy, as to render the evidence against him overwhelming to many minds. This, I have reason to know, has been the effect wrought, not only upon persons untrained in science, but also upon biologists of eminence both in this country and America. The state of medical opinion in England is correctly described in a recent number of the 'British Medical Journal,' where, in answer to the question, 'In what way is contagium generated and communicated?' we have the reply that, notwithstanding 'an almost incalculable amount of patient labour, the actual results obtained, especially as regards the manner of generation of contagium, have been most disappointing. Observers are even yet at variance whether these minute particles, whose discovery we have just noticed, and other disease-germs, are always produced from like bodies previously existing, or whether they do not, under certain favourable conditions, spring into existence *de novo*.'

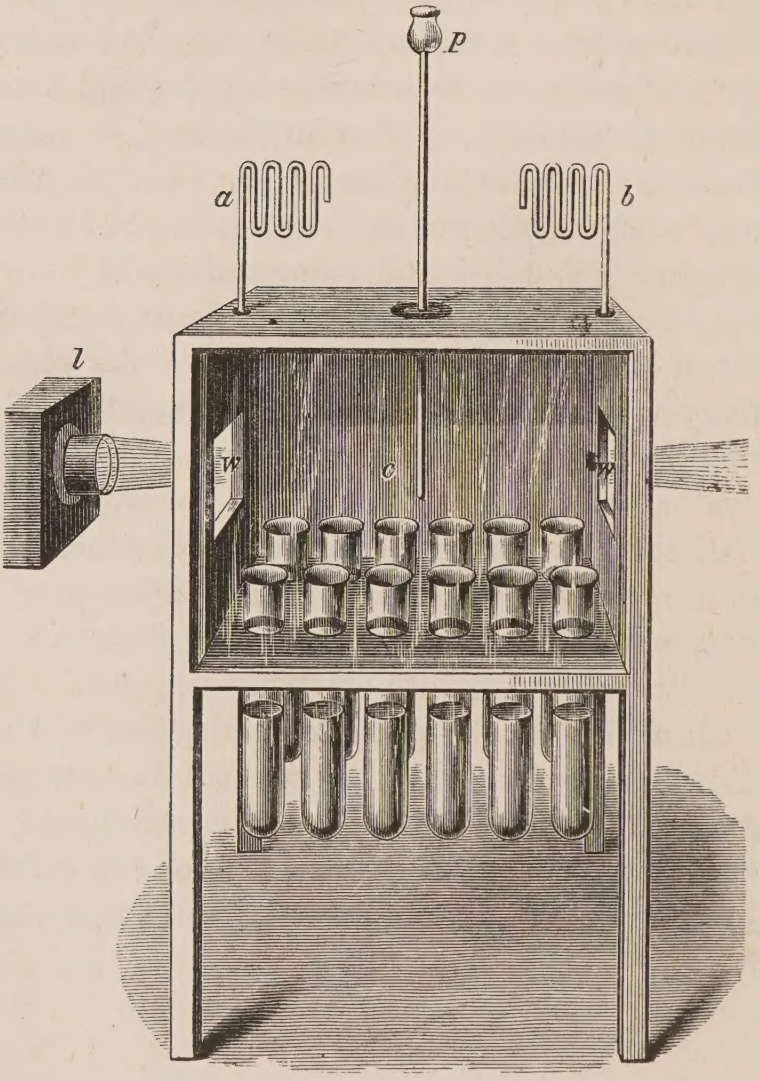
With a view to the possible diminution of the uncertainty thus described, I beg without further preface to submit to the Royal Society, and especially to those who study the etiology of disease, the following description of the mode of procedure followed in this inquiry, and of the results to which it has led.

§ 2. *Method of Experiment.*

A chamber, or case, was constructed, with a glass front, its top, bottom, back, and sides being of wood. At the back is a little door which opens and closes on hinges, while into the slides are inserted two panes of glass, facing each other. The top is perforated in the

middle by a hole 2 inches in diameter, closed air-tight by a sheet of india-rubber. This sheet is pierced in the middle by a pin, and through the pin-hole is passed the shank of a long pipette ending above in a small funnel.

FIG. 2.



A circular tin collar, 2 inches in diameter and $1\frac{1}{2}$ inch deep, surrounds the pipette, the space between both being packed with cotton-wool moistened by glycerine. Thus the pipette, in moving up and down, is not only firmly clasped by the india-rubber, but it also passes

through a stuffing-box of sticky cotton-wool. The width of the aperture closed by the india-rubber secures the free lateral play of the lower end of the pipette. Into two other smaller apertures in the top of the chamber are inserted, air-tight, the open ends of two narrow tubes, intended to connect the interior space with the atmosphere. The tubes are bent several times up and down, so as to intercept and retain the particles carried by such feeble currents as changes of temperature might cause to set in between the outer and the inner air.

The bottom of the box is pierced with two rows of holes, six in a row, in which are fixed, air-tight, twelve test-tubes, intended to contain the liquid to be exposed to the action of the moteless air.

The arrangement is represented in fig. 2, where *w w* are the side windows through which the searching beam passes from the lamp *l* across the case *c*; *p* is the pipette, and *a*, *b*, are the bent tubes connecting the inner and outer air. The test-tubes passing through the bottom of the case are seen below.

On the 10th of September, 1875, this case was closed. The passage of a concentrated beam across it through its two side windows then showed the air within it to be laden with floating matter. On the 13th it was again examined. Before the beam entered, and after it quitted the case, its track was vivid in the air, but within the case it vanished. Three days of quiet had sufficed to cause all the floating matter to be deposited on the interior surfaces, where it was retained by a coating of glycerine, with which these surfaces had been purposely varnished.

§ 3. *Deportment of Urine.*

The pipette being dipped into the tubes, fresh urine was poured into eight of them in succession on the 13th

of September. Each tube was about half-filled with the liquid. The tubes were then immersed in a bath of brine, raised to ebullition, and permitted to boil for five minutes. Aqueous vapour rose from the liquid into the chamber, where it was for the most part condensed, the uncondensed portion escaping, at a low temperature, through the bent tubes at the top. Before the brine was taken away little stoppers of cotton-wool were inserted in the bent tubes, lest the re-entrance of the air into the cooling-chamber should at first be forcible enough to carry motes along with it. As soon, however, as the outside temperature was assumed by the air within the case the cotton-wool stoppers were removed.

The front and back of this chamber were squares of 14 inches the side, the depth of the chamber being 8·5 inches. It contained, therefore, 1666 cubic inches of air, which had unimpeded access to the liquid in the tubes. No stoppers were employed. The air was unaffected by calcination, or even by filtering. Neither cotton-wool nor hermetic sealing was resorted to. Self-subsidence was the only means employed to rid the 'untortured' air of its floating matter.

A second series of eight tubes were filled at the same time with the same liquid, and subjected to the same boiling process. The only difference between the two series was, that these latter tubes were placed in a stand beside the case containing the former ones and exposed to the common air of the laboratory.

For the sake of distinction I will call the tubes opening into the case the *protected* tubes, and those opening into the common air the *exposed* tubes.

On the 17th of September all the protected tubes were bright and clear, while all the exposed tubes were distinctly turbid. Specks of mould, moreover, were in every case seen on the surface of the exposed liquid.

These waxed daily larger, and finally formed a thick layer on the top of every column. The liquid, meanwhile, changed from a pale sherry to a reddish-brown colour. To me the experiment was impressive in the highest degree.

On the 27th of September I provided myself with a microscope having a magnifying power of 1200 diameters. Under its scrutiny the turbidity of the liquid immediately resolved itself into swarms of *Bacteria* in active motion. Cohn correctly explains the turbidity. The index of refraction of the *Bacterium* being slightly different from that of the surrounding medium, a scattering of light is the consequence. This scattering, however, and the opalescence it produces, are practically independent of the motions of the *Bacteria*.

Since the date here referred to the exposed liquid has been frequently examined, both with the eye and with the microscope. To the former it is thickly turbid, to the latter it is swarming with life. Its smell is putrid. *All this time the protected tubes exhibit a liquid perfectly unchanged in appearance.* For four months it has remained as transparent and of as rich a colour as the brightest Amontillado sherry.

On the 1st of October another experiment similar in principle to that just described was begun. Fresh urine was employed, and a much smaller case. The capacity of the latter was 451 cubic inches ; and three test-tubes, instead of twelve, were passed air-tight through its bottom. Like those in the larger chamber they were filled by a pipette, and boiled for five minutes in a bath of brine. Beside them were placed three other tubes containing the same liquid treated in exactly the same way, but exposed to the common air. On the 5th all the exposed tubes were turbid, and found by microscopic examination to be swarming with *Bacteria*. The colour

of the exposed liquid had changed from a pale sherry colour to a brown orange. On the 25th the tubes were again examined, and found full of *Bacteria*. Two months subsequent to this latter date the infusion, diminished by evaporation, was found well charged with Bacterial life.

While this process of putrefaction was going on outside, the tubes opening into the moteless air of the case remained perfectly clear and void of life.

The large chamber represented in fig. 2, and above described, was the first operated on, and the liquid is shown by the draughtsman as filling only a small portion of the test-tubes. This smallness of volume is in part due to evaporation. Test-tubes 1·2 inch wide and 9 inches long were, in all subsequent experiments, nearly filled with the infusions. Strong in the first instance, these were sometimes kept until slow evaporation through the bent tubes at the top of the case had reduced them to one third or one fourth of their original volume. Each experiment, therefore, was, in reality, a series of experiments, extending over months, on infusions of different strengths, the concluding ones of the series attaining a very high degree of concentration.

§ 4. *Mutton-Infusion.*

A new case was constructed to contain six test-tubes. It, like the others, had a front of glass, side windows, and a back door. Its capacity was 857 cubic inches. It was sealed up on the 21st of September, and found free from floating matter on the 24th. The lean of mutton, cut into small pieces, was digested, or soaked, for four hours in water of a temperature of 120° F.[1] The infu-

[1] The temperature recommended by the supporters of spontaneous generation.

sion was then carefully filtered, and introduced into the six test-tubes by a pipette which was never removed from the case.

The mutton-juice was of a fine ruby colour; but on boiling, its albumen was precipitated, subsequently sank, and carried the colouring-matter with it. The supernatant liquid was perfectly clear. The frothing was considerable when the boiling began. Beside this new case was placed a stand containing six test-tubes filled with the same infusion, but exposed to the common air.

On the 27th all the outside flasks were perceptibly turbid; on the 28th they were found well filled with *Bacteria*, which on the 30th had increased to astonishing swarms. On the 15th of October the tubes were again examined, and found charged with undiminished life. They remained thus 'putrid' until the 14th of November.

During the whole of this time the infusion in contact with the moteless air of the chamber remained as clear as distilled water, and entirely free from life.

On the 14th of November I infected one of the clear tubes by introducing into it through the pipette a few drops of mutton-infusion which had been prepared and exposed upon the 12th of November, and which two days had sufficed to render turbid. On the 15th the inoculated infusion showed signs of turbidity, and on the 16th putrefaction had actively set in, the liquid being thickly muddy and full of life.

With a moteless chamber and three tubes, experiments were subsequently made on a second infusion of mutton. In this case, however, the infusion was boiled, its albumen was precipitated, and removed by filtration prior to its introduction into the chamber. The pellucid liquid was introduced on the 1st of October, boiled for

five minutes in the brine-bath, and abandoned to the air of the case. A series of exposed tubes containing the same infusion, similarly treated, was placed beside the protected ones. On the 4th all the outside tubes were muddy and swarming with *Bacteria*. Schroeder and Cohn have shown that different colours are produced by different kinds of *Bacteria*. In the three exposed tubes here referred to a yellow-green pigment was developed.

More than three months after its preparation, the infusion, considerably diminished by evaporation, remained in all the protected tubes as clear as at first.

§ 5. *Beef-Infusion.*

A beef-steak, after having its fat removed, was cut up into small pieces, and digested for three hours at a temperature of 120° F. The liquid was then poured off, boiled, and filtered. It was as clear and colourless as pure water. On the 4th of October it was introduced into three tubes protected by a chamber of 451 cubic inches capacity. It was boiled for five minutes in a brine-bath. Three exposed tubes, containing the same infusion, were placed beside the protected ones. On the 5th the exposed tubes showed signs of haziness, on the 6th they were turbid, of a green colour, and filled with *Bacteria*. They have maintained for months their muddiness, colour, and swarming life.

While the exposed beef-infusion putrified in this way, all the protected infusions remained perfectly sweet and clear.

§ 6. *Haddock-Infusion.*

The haddock was cut up and digested on the 24th of September; it was afterwards introduced into six tubes, protected by a chamber. On boiling, its albumen,

like that of the mutton first referred to, coagulated and sank to the bottom, leaving a perfectly clear liquid overhead. Six exposed tubes filled with the same infusion were placed beside the six protected ones.

On the 27th the exposed tubes were all turbid and swarming with *Bacteria*. On the 29th one of the tubes showed a fine green colour; three other tubes showed the same colour afterwards. The vivacity of the organisms was extraordinary, and their shapes various. They darted rapidly to and fro across the field, clashing, recoiling, and pirouetting—rendering it, indeed, difficult to believe in the vegetable nature which the best microscopists assign to them.

For nearly three weeks the protected tubes remained perfectly clear. To gain room, the case was subsequently shifted, and soon afterwards one of the six tubes became turbid with organisms, the germs of which had obviously been shaken into the tube.

For more than a month this single infected flask remained in company with the five healthy ones. The air containing the gaseous products of putrefaction had free access to the whole of them, but there was no spread of the infection. As long as the organisms themselves were kept out of the flasks, the 'sewer-gas' developed by the putrefaction had no infective power. On the 14th of November I infected two of the five perfectly pellucid tubes with haddock-infusion which, after boiling, had been exposed for two days to the air. On the 15th the two tubes had obviously yielded to the infection. On the 16th disease, if I may use the term, had completely taken possession of them. Into one of them only one or two drops of the turbid infusion had fallen, while ten times this amount was introduced into the other. Nevertheless on the 16th both appeared equally turbid. The infection acted exactly like the virus of

smallpox, a small quantity of which will in the long run produce the same effect as a large one.

§ 7. *Turnip-Infusion.*

Turnip-juice had a special interest for me in consequence of the important part it plays in the experiments of heterogenists. I turned to it with the anxious desire to learn whether the statements made concerning it were correct.

The conditions laid down as to the strength of the infusion, the temperature to be maintained during the process of digestion, and the time it was to be maintained[1] were scrupulously adhered to. Thus the turnip was cut into thin slices, and digested for four hours in a beaker of water immersed in a water-bath kept at a temperature close to 120° Fahr. The infusion was then carefully filtered, introduced through a pipette into its case, and boiled there for five minutes. Six protected test-tubes were charged with the infusion on the 24th of September, while six other tubes were placed on a stand outside, and exposed to the common air of the laboratory.

On the 27th the exposed tubes were distinctly turbid, and on microscopic examination were found peopled with *Bacteria*. The protected tubes, on the contrary, were perfectly clear. A little distilled water had been added to one of the outer tubes. The germinal matter, whatever it may be, must have been copious in the water; for the tube to which it was added far exceeded the other two in the rapidity of life-development. On the 30th this tube contained *Bacteria* in swarms, of small size, but of astonishing activity. The other tubes also were fairly charged with organisms,

[1] Bastian, 'Beginnings of Life,' vol. i. p. 357, note.

larger and more languid, but not at all so numerous as in the watered tube. On the 5th of October some of the exposed tubes began to clear; as if the *Bacteria* had died through lack of nutriment, and were falling as a thick sediment to the bottom.

During these changes the protected tubes were visibly unaltered, the liquid within every one of them remaining as clear as it had been on the day of its introduction.

In this instance I was specially anxious to verify the result by repetition. Two other cases were therefore fitted up to contain three tubes each, and instead of a door a movable panel was placed at the back. After two or three days' rest both cases were found free from floating matter, and on the 1st of October the turnip-infusion was introduced, and boiled for five minutes in a bath of brine.

In the former experiment the temperature of digestion was maintained by keeping the beaker containing the turnip in a bath of warm water. In the present instance the turnip was sliced in a dish and placed before a fire. An occult but efficient power like that already ascribed to the actinic rays[1], might, I thought, be ascribed to radiant heat, and I therefore copied to the letter the mode of digesting pursued by modern heterogenists.

Adjacent to the closed cases was placed a series of three exposed tubes, containing a liquid prepared in precisely the same way. On the 4th of October the exposed tubes were all turbid, and swarmed with *Bacteria*. In two of the tubes they were distinctly more numerous and lively than in the third. Such differences between sensibly conterminous tubes, containing the same infusion, are frequent. On the 9th, moreover, the two most

[1] 'Nature,' vol. iii. p. 247.

actively charged tubes were in part crowned by beautiful tufts of *Penicillium glaucum*.[1] This expanded gradually until it covered the entire surface with a thick tough layer, which must have seriously intercepted the oxygen necessary to the Bacterial life. The *Bacteria* lost their translatory power, fell to the bottom, and left the liquid between them and the superficial layer clear.

Another difference, pointing to differences in the unseen life of the air, was shown by these tubes. The turbidity of the two mould-crowned ones was colourless, exhibiting a grey hue. The third tube, the middle one of the three, contained a bright yellow-green pigment, and on its surface no trace of mould was to be seen. It never cleared, but maintained its turbidity and its Bacterial life for months after the other tubes had ceased to show either. It cannot be doubted that the mould-spores had fallen into this tube also, but in the fight for existence the colour-producing *Bacteria* had the upper hand. Six other tubes, similarly exposed, showed the grey muddiness: all of them became thickly covered with mould, under which the *Bacteria* died or passed into a quiescent state, fell to the bottom, and left the liquid clear.

Up to the 13*th of October the purity of the six protected tubes remained unimpaired.*

Here a complementary experiment was made. It remained to be proved that those long-dormant clear infusions had undergone no change which interfered with their ability to develop and maintain life. On the 13th of October, therefore, the small panel was removed from the back of one of the cases, and with three new pipettes specimens were taken from the three tubes within it. The closest search revealed no living thing.

[1] Ordinary mould.

The air of the laboratory being permitted to diffuse freely into the case, on the day after the removal of the panel the test-beam showed the case to be charged with floating matter.

The access of this matter was the only condition necessary to the production of life; for on the 17*th all the tubes were muddy and swarming with* Bacteria.

A similar experiment, subsequently made, revealed some of the snares and pitfalls which await an incautious worker. The chamber already referred to as containing six tubes, filled with turnip-juice, preserved the infusion clear for a month. On the 21st of October the back door of the chamber was opened, and specimens of the clear infusion were taken out for examination by the microscope. The first tube examined showed no signs of life. This result was expected. Picking up another pipette, I took a sample from the second tube. Here, to my astonishment, the exhibition of life was monstrously copious. There were numerous globular organisms, which revolved, rotated, and quivered in the most extraordinary manner. There were also numbers of lively *Bacteria* darting to and fro. An experimenter who ponders his work and reaches his conclusions slowly, cannot immediately relinquish them: and in the present instance some time was required to convince me that no mistake had been made. I could find none, and was prepared to accept the conclusion that in the boiled infusion, despite its clearness, life had appeared.

But why, in the protected turnip-infusion, which had been examined on the 13th of October, could no trace of life be found? In this case perfect transparency was accompanied by an utter absence of life. The selfsame action upon light that enabled the *Bacteria* to show themselves in the microscope must, one would

think, infallibly produce turbidity. Why, moreover, should life be absent from the first member of the present group of tubes? I searched this again, and found in it scanty but certain signs of life. This augmented my perplexity. A third tube also showed traces of life. I reverted to the second tube, where life had been so copious, and found that in it the organisms had become as scanty as in the others. I confined myself for a time to the three tubes of the first row of the six, going over them again and again; sometimes finding a *Bacterium* here and there, but sometimes finding nothing. The first extraordinary exhibition of life it was found impossible to restore. Doubtful of my skill as a microscopist, I took specimens from the three tubes and sent them to Prof. Huxley, with a request that he would be good enough to examine them.

On the 22nd the search was extended to the whole of the tubes. Early in the day lively *Bacteria* were found in one of them; later on, not one of the six yielded to my closest scrutiny any trace of life. On the evening of the 22nd a note was received from Prof. Huxley stating that a careful examination of the specimens sent to him revealed no living thing.

Pipettes had been employed to remove the infusion from the test-tubes. They were short pieces of narrow glass tubing, drawn out to a point, with a few inches of india-rubber tubing attached to them. This was found convenient for bending so as to reach the bottom of the test-tubes. Suspicion fell upon this india-rubber. It was washed, the washing-water was examined, but no life was found. Distilled water had been used to cleanse the pipettes, and on the morning of the 23rd I entered the laboratory intending to examine it. Before dipping a pipette into the water I inspected its point.

The tiniest drop had remained in it by capillary attraction from the preceding day. This was blown on to a slide, covered, and placed under the microscope. An astonishing exhibition of life was my reward. Thus on the scent, I looked through my pipettes, and found two more with the smallest residual drops at the ends: both of them yielded a field rampant with life. The *Bacteria* darted in straight lines to and fro, bending right and left along the line of motion, wriggling, rotating longitudinally, and spinning round a vertical transverse axis. Monads also galloped and quivered through the field. From one of these tiny specks of liquid was obtained an exhibition of life not to be distinguished from that which had astonished me on the 21st.

Obviously the phenomenon then observed was due to the employment of an unclean pipette. Equally obvious is it that in inquiries of this nature the experimenter is beset with danger, the grossest errors being possible when there is the least lack of care.

The chamber here operated on had been opened with a view to testing the capacity of the infusions within it to develop and maintain life. *For four weeks they had remained perfectly clear.* Two days after the door was opened and the common laboratory air admitted all six tubes were turbid, and swarming with *Bacteria.* Some of them were very long, and their wriggling and darting hither and thither very impressive.

The same chamber was again thoroughly cleaned, sealed, and permitted to remain quiet until the floating matter had subsided. On the 17th of November a fresh infusion of turnip was introduced into it through the pipette, boiled in an oil-bath, and again abandoned to the air of the case.

After several months the infusion in every tube of the six remained as clear as it was on the day of its introduction.

Six other tubes charged with the same infusion, boiled in the same way, became turbid in a few days, and subsequently covered with thick layers of *Penicillium*.

§ 8. *Hay-Infusion.*

This infusion has been credited with a power of spontaneous generation similar to that ascribed to turnip-juice. The hay being chopped into short lengths was digested for four hours in water kept at a temperature of 120° Fahr. On the 24th of September the filtered infusion was introduced into its chamber, and boiled there for five minutes. Six tubes were charged with the protected liquid, while six other tubes, filled with the same infusion, were placed on a stand outside the case.

On the 27th the inside flasks were clear, the outside ones faintly turbid. On the 28th spots of mould appeared upon all the exposed surfaces. The infusion in one of the outside tubes had been diluted with distilled water, and in it the development of life was far more rapid than in the five others; all of them, however, on the 28th contained *Bacteria*.

On the 29th I noticed a larger organism than the *Bacteria* moving rapidly to and fro across the field, the drop containing it being taken from the dilute infusion. Several of these monads were seen upon the 30th gambolling among the smaller *Bacteria*, appearing bright or dark as they sank or rose in the liquid, a film of which, large as they looked, was to them an ocean. Swarms of *Bacteria* were seen on the 2nd of October, their translatory motions being so rapid and varied, and so apparently guided by a purpose, as to render it difficult to believe that they could be anything else than animals. On the 15th there was a marvellous exhibition of the larger

Infusoria, which appeared to have driven the *Bacteria* from their habitat, as few of the latter were to be seen. My inability to find the larger creatures a second time in such numbers perplexed me, causing me to conclude that I had accidentally alighted upon a colony of them. My experience with the unclean pipettes already described, pointed, however, to another source.

While three days sufficed to break down their purity, and to fill the six exposed tubes with Bacterial life, *the six protected ones remained for more than three months as clear and healthy as they were on the day the infusion was poured into them.* Neither a trace of mould upon the surface of any one of them, nor a trace of turbidity in its mass, was to be seen.

Into another chamber containing three test-tubes a very strong infusion of hay was introduced on the 1st of October. It was boiled for five minutes, and then abandoned to the air of the case. Three other tubes exposed to the laboratory air were placed on a stand beside the case. The colour of the infusion was very deep, but it was quite transparent. One of the outer tubes was diluted with distilled water. On the 3rd the infusion in this tube was turbid, the other exposed ones remaining clear. The unseen germinal matter had in some way or other invaded the distilled water, and made it infective. The dilute infusion contained multitudes of *Bacteria*, many motionless, but many moving rapidly about. On the 4th of October all the tubes swarmed with *Bacteria*. They continued muddy till the middle of November, when they were employed for experiments on infection.

Throughout the whole of this time the protected tubes remained unchanged.

With regard to infection, it may be stated here that

the merest speck of a vegetable infusion containing *Bacteria* infects all animal infusions, and *vice versâ*. The bursting of a bubble infects an infusion reached by the spray. It is the envelope, and not the gas of the bubble, which produces this result.

Other experiments on hay-infusions, acid, neutral, and alkaline, placed in contact with air purified in various ways, yielded in 1875 the same negative result.

§ 9. *Infusion of Sole.*

The fish was cut up and digested for three hours in water kept at 120° Fahr. On the 17th of November it was introduced into a case containing three test-tubes, and boiled there for five minutes. Three other tubes hung outside the case were exposed to the ordinary laboratory air.

The three exposed tubes were feebly but distinctly cloudy on the 19th. On the 22nd they were all thickly turbid. Scattered spots of *Penicillium* then appeared on two of them, while the third tube, which stood between these two, kept the *Penicillium* down. This central tube contained the pigment-forming *Bacteria*, which have frequently shown a singular power in preventing the development of mould. For nearly two months the central tube successfully withstood this development, while its two neighbours were covered by a matted layer of *Penicillium*.

During the whole of this time the protected infusion continued as clear and colourless as distilled water.

§ 10. *Liver-Infusion.*

On the 10th of November the infusion was prepared by the process of digesting already so often described.

It was introduced into a case containing three protected tubes, and boiled there for five minutes in the brine-bath. Hung on to the chamber at the same time were three tubes containing the same infusion, but exposed to the common air. On the 13th *Bacteria* were numerous in the exposed tubes, and soon afterwards all three of them became thickly muddy and putrescent. They continued so for months.

The protected tubes, on the contrary, showed throughout a bright yellow liquid, as transparent and fresh as it was on the day of its introduction into the case.

§ 11. *Infusions of Hare, Rabbit, Pheasant, and Grouse.*

For the sake of economy, as so many of them were employed, the shape of the cases was subsequently varied. The rounded end of a tall glass shade was cut off, so as to convert the shade into a hollow cylinder, open at both ends. This was set upright on a wooden stand, and cemented to it air-tight. Through the stand passed three large test-tubes also air-tight. To the top of the cylinder was cemented a circular piece of wood, the middle of which was occupied by a pipette passing first through india-rubber and then through a stuffing-box of cotton-wool moistened by glycerine.[1] The air within the case was connected with the air without by means of the open bent tubes already described.

In the first experiments made with these cases defects of construction were revealed during the boiling of the infusions. But increased experience enabled me to

[1] In the earlier experiments the india-rubber formed the bottom of the stuffing-box, where particles were sometimes detached from it by the motion of the pipette. To prevent this the positions of wool and rubber were afterwards reversed.

render them secure. The floating matter within the cases having been permitted to subside, into four of them, on the 30th of November, infusions of hare, rabbit, pheasant, and grouse were introduced. The infusions were boiled in the usual way, and abandoned to the air of the case. Outside each case, and hung on to it, were three test-tubes of the same size and containing the same infusion as that within.

Examined on Christmas-day, the following were the observed results :—

Pheasant.—The three interior tubes perfectly limpid : the three exposed ones turbid and covered with *Penicillium.*

Grouse.—In the same condition as the pheasant.

Hare.—The same as grouse and pheasant.

Rabbit.—The three interior tubes covered with tufts of particularly beautiful *Penicillium*, some of the tufts striking deep into the liquid. In two out of the three tubes, moreover, mycelium was flourishing below. All the outer tubes were, as usual, turbid and covered with *Penicillium.*

Is this, then, a case of spontaneous generation ? Without further evidence no cautious worker would draw such a conclusion. Opposed to this isolated instance stand all the others mentioned in these pages, and their proper action on the mind is to compel it to demand the closest scrutiny before accepting this apparent exception as a real one. Subjected to such scrutiny, it appeared that of the four shades the one containing the rabbit-infusion, and that only, had yielded to the heat of boiling. The shade had been fastened upon its slab with plaster and cement, which became so loose during the boiling that the steam issued from the chinks. But crannies which could permit steam to escape could permit air to enter, and to

the presence of such air the appearance of the *Penicillium* was doubtless due.

I did not, however, rest content with mere inference, but tested the rabbit-infusion by placing three fresh tubes of it in one of the firmer cases first described. It was introduced and boiled on the 5th of January, 1876, three other tubes filled with the same boiled infusion being exposed on the same day to the ordinary air. The three protected tubes remained clear for three months, while in three days the three exposed ones were charged with *Bacteria*.

Salmon.—The colouring-matter of this fish did not at all affect the infusion; indeed, no better example of original freedom from colour or opalescence, and of persistent purity in contact with the moteless air, has occurred to me than salmon-infusion. It was introduced into a cylindrical case on the 13th of December, where it continued for months to show the brilliant transparency exhibited at first. Three unprotected tubes, on the other hand, became turbid and covered with mould in a few days.

Hops.—One tube of this infusion was protected simply by a lamp-glass, corked and cemented above and below. Through the lower cork passed the single test-tube, air-tight; while through the upper one passed the pipette and the bent tubes intended to connect the outer and the inner air. The infusion was prepared and introduced on the 28th of October. In a few days the exposed tube was found turbid and covered with mould; the protected tube, on the contrary, remained clear for several months.

Tea and Coffee.—One tube of each was protected by a lamp-glass similar to that employed in the infusion of hops. Both were prepared on the 28th of October, exposed tubes being hung up at the same time. The

protected tea has remained clear, while the exposed tea is turbid and covered with mould. Both the exposed coffee and the protected coffee are, on the other hand, turbid and covered with *Penicillium*.

The remarks already made with regard to the rabbit-infusion apply here. The case is one, not for the hasty admission of spontaneous generation, but for further scrutiny. I examined the apparatus as it stood. The pipette used to introduce the coffee (and this one only of the three employed in these experiments) rested against the outer edge of the tube containing the infusion. This had in part evaporated, had been in part re-condensed, and had trickled down the pipette so as to form a small drop at the point where pipette and tube touched each other. The drop had virtually washed the outer surface of the pipette, carrying with it, in part, such matter as might have attached itself to that surface. A portion of this washing-water reaching the infusion was clearly the origin of the life observed. The sure test, however, was the repetition of the experiment under conditions which should exclude this source of error. On the 27th of December accordingly two tubes protected by lamp-glasses were prepared, two other tubes of the infusion being exposed to the air. The former remained clear for months, the latter in the same number of days became turbid and covered with *Penicillium*.

§ 12. *Infusions of Codfish, Turbot, Herring, and Mullet.*

With a view of causing these experiments on moteless and mote-laden air to run parallel with others made with hermetically-sealed tubes, to be described further on, I added the fish named in the heading of this section to the other substances examined. The mullet was in-

troduced into its case on the 3rd of January. The warm air of the room had, however, so acted on the wood of the chamber, which had been employed in former experiments, that the water of condensation trickled from a chink in the bottom. The other chambers were mended as far as possible, and into them the infusions were introduced on the 4th of January. Each chamber, as before, was provided with three exposed tubes for comparison with three protected tubes within. On the morning of the 6th the exposed turbot-infusion was clear in all the tubes; a few hours subsequently two out of the three became cloudy; while on the 7th *Bacteria* had taken possession of all of them. All the unprotected tubes of codfish were cloudy on the 6th, more cloudy on the 7th, and covered with a soapy layer upon the 8th. The three exposed herring-tubes were also cloudy on the 6th, the cloudiness advancing afterwards to thicker turbidity. The mullet gave way in the same manner. *For more than three months the protected tubes, including even the imperfect chamber which protected the mullet-infusion, have remained as clear as they were upon the day of their introduction.*

To these fish-infusions may be added others of eel and oyster. Two tubes of each, protected by lamp-glasses, were charged on the 27th of December. They remain unchanged. Two other pairs of tubes, prepared in the same way and exposed to the laboratory air, are turbid and covered with *Penicillium*.

§ 13. *Infusions of Fowl and Kidney.*

Three tubes of the fowl-infusion were introduced into a case, and boiled there for five minutes, on the 4th of January. Three similar tubes were at the same time exposed to the air. On the 6th all the outer

tubes were cloudy, the cloudiness becoming denser on the following days, while disks of *Penicillium* began to form on the exposed surfaces. It was found exceedingly difficult to obtain a clear infusion of kidney. The liquid, after it had passed through a dozen filters, was still quite muddy. With considerable labour and care, and by the employment of 200 filters, the mechanically suspended matter was at length removed, and a clear infusion obtained. It was introduced into its case, to which three exposed tubes were attached, on the 4th of January. On the 7th the latter were perceptibly cloudy, on the 8th distinctly so, while specks of mould rested upon them all. The protected tubes, on the contrary, have for months maintained their transparency undimmed.[1]

The entire number of experiments made to illustrate the association of scattered light and Bacterial and fungoid life are not here recounted. Whiting, for example, may be added to the fish, and pork to the flesh examined, while many of the other substances have been tested oftener than I have thought it necessary to record. The method of boiling was also varied in a manner which may claim a passing reference here.

§ 14. *Boiling by an Internal Source of Heat.*

Two large test-tubes were fixed air-tight in the same case. On the 8th of November, after the floating matter had subsided, infusions of hay and turnip were introduced. Dipping deep into each infusion were two tinned copper wires, connected below by a spiral of

[1] Kidney has been mentioned by Dr. Bastian as a substance with which he demonstrates the occurrence of spontaneous generation. He does not mention the extraordinary turbidity of the infusion, which proved so troublesome to me.

platinum wire. The copper wires passed through the case, and were connected with a voltaic battery outside. The spiral was heated by the current from this battery. After a few minutes' heating ebullition set in, and was continued for five minutes in each tube. Two other tubes containing the same infusions were boiled in the same way, and afterwards hung outside the case containing the two protected tubes.

In another chamber were placed two tubes containing infusions of beef and mutton. The arrangement and the treatment were precisely the same as those just described in the case of hay and turnip.

Examined some months subsequently, the exposed tubes of all four infusions were found turbid and covered with *Penicillium*, while all the four protected tubes remained unchanged. During the boiling process some flocculi detached themselves from the tinned surfaces of the copper wires; but in the protected tubes these have fallen to the bottom, and left the supernatant liquid clear. Platinum wires would have been better than tinned copper ones.

§ 15. *Partial Discussion of the Results.*

Thus by experiments, reiterated in many cases, with urine, mutton, beef, pork, hay, turnip, tea, coffee, hops, haddock, sole, salmon, codfish, turbot, mullet, herring, eel, oyster, whiting, liver, kidney, hare, rabbit, fowl, pheasant, grouse, has the induction been established, that the power of developing Bacterial life by atmospheric air and its power of scattering light, go hand in hand. We shall immediately examine more closely what this means.

The necessity of employing strong infusions has been frequently dwelt upon, if we would realize the

phenomena of spontaneous generation. I would therefore recall to mind what has been stated on a previous page, that in most of the experiments here described the infusion at starting was strong, and that it was permitted to evaporate with extreme slowness until its concentration became three or four fold what it had been at starting. Every experiment was thus converted into an indefinite number of experiments on infusions of different strengths. Never, in my opinion, was the requirement as to concentration more completely fulfilled, and never was the reply of Nature to experiment more definite and satisfactory. The temperatures, moreover, to which the infusions have been subjected embrace those hitherto alleged to be effectual, extending indeed beyond them in both directions.[1] They reached from a lower limit of 50° to a higher limit of more than 100° Fahr. Still higher temperatures were applied in other experiments to be described subsequently. With regard to the number of the infusions, more than fifty moteless chambers, each with its system of tubes, have been tested. There is no shade of uncertainty in any of the results. In every instance we have, within the chamber, perfect limpidity and sweetness—without the chamber, putridity and its characteristic smells. In no instance is the least countenance lent to the notion that an infusion deprived by heat of its inherent life, and placed in contact with air cleansed of its visibly suspended matter, has any power whatever to generate life anew.

If it should be asked how I have assured myself that the protected liquids do not contain *Bacteria*, I would, in the first place, reply that with the most careful microscopic search I have been unable to find

[1] See Proc. of Roy. Soc. vol. xxi. p. 130, where a temperature of 70° is described as effectual.

them. But much more than this may be affirmed. The electric or the solar beam is a far more powerful and searching test in this matter than the microscope. In the foregoing pages I have more than once described the clearness of my protected infusions, after months of exposure, as equal to that of distilled water. So far is this from being an exaggeration, that it falls short of the truth; for I have never seen distilled water so free from suspended particles as the protected infusions prove themselves to be. When for months a transparent liquid thus defies the scrutiny of the searching beam, maintaining itself free from every speck which could scatter light as a *Bacterium* scatters it—when, moreover, an adjacent infusion, prepared in precisely the same way, but exposed to the ordinary air, becomes first hazy, then turbid, and ends by wholly shattering the concentrated beam into irregularly scattered light, I think we are entitled to conclude that *Bacteria* are as certainly absent from the one as they are present in the other. (See Note I. at the end of this paper.)

For the right interpretation of scientific evidence something more than mere sharpness of observation is requisite, very keen sight being perfectly compatible with very weak insight. I was therefore careful to have my infusions inspected by biologists, not only trained in the niceties of the microscope, but versed in all the processes of scientific reasoning. Their conclusion is that it would simply weaken the demonstrative force of the experiments to appeal to the microscope at all.

§ 16. *Suspended Particles in Air and Water; their relation to* Bacteria.

Examined by the concentrated solar rays, or by the condensed electric beam, the floating matter of the air

is seen to consist:—first, of particles so coarse that their individual motions can be followed by the eye; secondly, of a finer matter which is not to be distinguished as motes, but which emits a uniform and changeless light. In this finer matter the coarser motes move as in a medium.

As regards the production of colour, the action of small particles has been examined by Brücke in a paper 'On the Colours of Turbid Media.'[1] In relation to the question of polarization, Professor Stokes has made some remarks in his memoir 'On the Change of the Refrangibility of Light.'[2] I may also be permitted to refer to my own papers 'On New Chemical Reactions by Light' and 'On the Blue Colour of the Sky,' in the Proceedings of the Royal Society for 1868–69, and to a paper 'On the Action of Rays of High Refrangibility on Gaseous Matter,' in the Philosophical Transactions for 1870. M. Soret, Lord Rayleigh, and Mr. Bosanquet have also worked at this subject, which, as far as it now concerns us, a few words will render clear.

When the track of a parallel beam in dusty air is looked at horizontally through a Nicol's prism, in a direction perpendicular to the beam, the longer diagonal of the prism being vertical, a portion of the light from the finer matter, being polarized, is extinguished. The coarser motes, on the other hand, which do not polarize the light, flash out with greater force, because of the increased darkness of the space around them.

The individual particles of the finest floating matter of the air lie probably far beyond the reach of the microscope. At all events it is experimentally demonstrable that there are particles which act similarly upon light, and which are entirely ultra-microscopic. A few

[1] Pogg. Ann. lxxxviii. p. 363.

[2] Philosophical Transactions, vol. 142, pp. 529–530.

days ago, for example, an inverted bell-jar was filled with distilled water, into which, while it was briskly beaten by a glass rod, was dropped a solution of mastic in alcohol. The proportion was less than that employed by Brücke, being about ten grains of the gum to 1,000 grains of the alcohol. The jar was placed under a skylight, at the height of the eye above the floor. It was of a beautiful cerulean hue, this colour arising wholly from the light scattered by the mastic particles. Looked at horizontally through a Nicol's prism, with its shorter diagonal vertical, the blue light passed freely to the eye. Turning the long diagonal vertical, the scattered light was wholly quenched, and the jar appeared as if filled with ordinary pure water.

I tried the effect of a powerful filter upon those particles, and found that they passed sensibly unimpeded through forty layers of the best filtering-paper.[1]

The liquid containing them was examined by a microscope magnifying 1,200 diameters. The suspended mastic particles entirely eluded this power, the medium in which they swam being as uniform as distilled water in which no mastic whatever had been precipitated.

The optical deportment of the floating matter of the air proves it to be composed, in part, of particles of this excessively minute character. The concentrated beam reveals them collectively, long after the microscope has ceased to distinguish them individually. In London rooms, moreover, they are for the most part organic particles, which may be removed from the air by combustion. In presence of such facts, any argument against atmospheric germs, based upon their being beyond the reach of the microscope, loses all validity.

We are here brought face to face with a question

[1] There are filters, however, which stop them; but of this immediately.

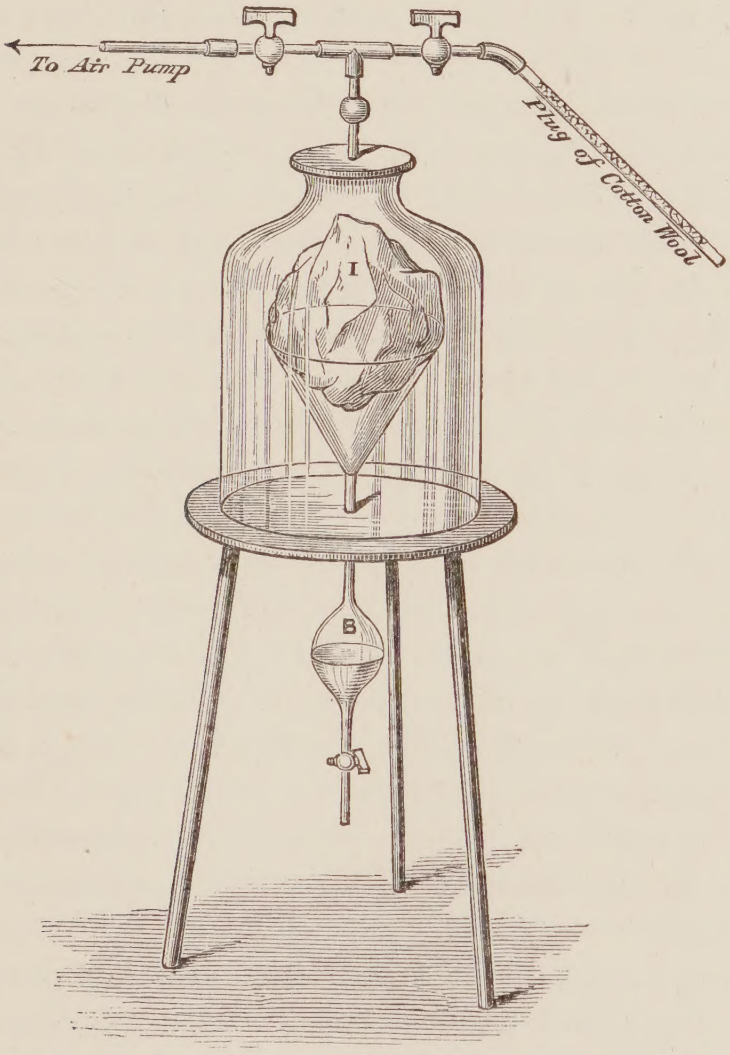
of extreme importance, which it will be useful to clear up. 'Potential germs' and 'hypothetical germs' have been spoken of with scorn, because the evidence of the microscope as to their existence was not forthcoming. Sagacious writers had drawn from their experiments the perfectly legitimate inference that in many cases the germs exist, though the microscope fails to reveal them. Such inferences, however, have been treated as the pure work of the imagination, resting, it was alleged, on no real basis of fact. But in the concentrated beam we possess what is virtually a new instrument, exceeding the microscope indefinitely in power. Directing it upon media which refuse to give the coarser instrument any information as to what they hold in suspension, these media declare themselves to be crowded with particles —not hypothetical, not potential, but actual and myriad-fold in number—showing the microscopist that there is a world far beyond his range.

In §§ 6 and 8 experiments on the infection of clear infusions by others containing visible *Bacteria* are referred to. But for the infection to be sure it is not necessary that the *Bacteria* should be visible. Over and over again I have repeated the experiments of Dr. Burdon Sanderson on the effective power of ordinary distilled water, in which the microscope fails to reveal a *Bacterium*. The water, for example, furnished to the Royal-Institution laboratory by Messrs. Hopkin and Williams is sensibly as infectious as an infusion swarming with *Bacteria*. The vessels are the source of infection here.

Perhaps the severest experiment of this kind ever made was one executed by Dr. Sanderson with water prepared by myself. In 1871 I sought anxiously and assiduously for water free from suspended particles. The liquid was obtained in various degrees of purity, but never entirely pure. Knowing the wonderful power of

extrusion, as regards foreign matter, brought into play by water in crystallizing, the thought occurred to me of examining the liquid derived from the fusion of the most transparent ice. At my request, therefore, my assistant arranged the following apparatus for me:—

Fig. 3.



Through the plate of an air-pump (fig. 3) passed air-tight the shank of a large funnel. A small glass bulb, B, furnished with a glass stopcock, was attached to the shank of the funnel below. Prior to being put together all parts of the apparatus had been scrupulously cleansed.

In the funnel was placed a block of ice, I, selected for its transparency, having a volume of 1000 cubic inches or thereabouts, and over the ice was placed an air-tight receiver. Several times in succession the air was removed from this receiver, its place on each occasion being taken by other air carefully filtered through cotton-wool. The transparent ice was thus surrounded by moteless air.

The ice was now permitted to melt; its water trickled into the small glass bulb below, which was filled and emptied a great number of times. From the very heart of the block of ice the water was finally taken and subjected to the scrutiny of the concentrated beam. It proved to be the purest liquid I had ever seen—probably the purest human eye had ever seen; but still it contained myriads of ultra-microscopic particles. The track of the beam through it was of the most delicate blue, the blue light being perfectly polarized. It could be wholly quenched by a Nicol's prism, the beam then passing through the liquid as through a vacuum. A comparison of the light with that scattered by mastic particles such as those above referred to, proved the suspended particles of the ice-water to be far smaller than those of the mastic. No microscope, therefore, could come near them.[1] Such water, however, was proved by Dr. Sanderson to be as infectious as the water from any ordinary tap.

Infinitesimal as these particles are, however, they may be separated by mechanical means from the liquid in which they are held in suspension. Filters of porous earthenware, such as the porous cells of Bunsen's battery,

[1] I have endeavoured to convey some notion of the smallness of these scattering particles in 'Fragments of Science,' Art. 'Scientific Use of the Imagination.' See note on Mr. Dallinger's observations at the end of this Memoir.

have been turned to important account in the researches of Dr. Zahn, Professor Klebs, and Dr. Burdon Sanderson. In various instances it has been proved that, as regards the infection of living animals, the porous earthenware intercepts contagia. For the living animal, organic infusions, or Pasteur's solution, may be substituted. Not only are ice-water, distilled water, and tap-water thus deprived of their powers of infection, but, by plunging the porous cell into an infusion swarming with Bacterial life, exhausting the cell, and permitting the liquid to be slowly driven through it by atmospheric pressure, the filtrate is not only deprived of its *Bacteria*, but also of those ultra-microscopic germs which appear to be as potent for infection as the *Bacteria* themselves. The precipitated mastic particles before described, which pass unimpeded through an indefinite number of paper filters, are wholly intercepted by the porous cell.

These germinal particles abound in every pool, stream, and river. All parts of the moist earth are crowded with them. Every wetted surface which has been dried by the sun or air contains upon it the particles which the unevaporated liquid held in suspension. From such surfaces they are detached and wafted away, their universal prevalence in the atmosphere being thus accounted for. Doubtless they sometimes attach themselves to coarser particles, organic and inorganic, which are left behind along with them ; but they need no such rafts to carry them through the air, being themselves endowed with a power of flotation commensurate with their extreme smallness and the specific lightness of the matter of which they are composed.

I by no means affirm that the developed *Bacterium*, which requires for its maintenance nutriment beyond that which ordinary water can always supply, is never

wafted through the air. Cases may arise favourable to the growth and dispersion of the full-grown organism. Whether, after desiccation, it retains the power of reproduction is another question. But it ought, I think, to be steadily borne in mind that the complete *Bacteria* and the atmospheric matter from which they spring are, in general, different things. I have carefully sought for atmospheric *Bacteria*, but have never found them. They have never, to my knowledge, been found by others; and that they arise from matter which has not yet assumed the Bacterial form is, as just shown, capable of demonstration. An organic infusion, boiled and shielded from atmospheric particles, will remain clear for an indefinite period, while a fragment of glass which has been exposed to the air, but on which no trace of a *Bacterium* is to be found, will in two or three days develop in the infusion a multitudinous crop of life.

We have now to look a little more closely at these particles, foreign to the atmosphere but floating in it, and proved beyond doubt to be the origin of all the Bacterial life which our experiments have thus far revealed. We must also look at them as they exist in water, in countless multitudes, being as foreign to this medium as the floating atmospheric dust is to the air in which it swims. The existence of the particles is quite as certain as if they could be felt between the fingers, or seen by the naked eye. Supposing them to augment in magnitude until they come, not only within range of the microscope, but within range of the unaided senses. Let it be assumed that our knowledge of them under these circumstances remains as defective as it is now—that we do not know whether they are germs, particles of dead organic dust, or particles of mineral matter. Suppose a vessel (say a flower-pot) to be at hand filled with nutritious earth, with which we mix our unknown

particles, and that in forty-eight hours subsequently buds and blades of well-defined cresses and grasses appear above the soil. Suppose the experiment when repeated a hundred times to yield the same unvarying result. What would be our conclusion? Should we regard those living plants as the product either of dead dust, or of mineral particles? or should we regard them as the offspring of living seeds? The reply is unavoidable. We should undoubtedly consider the experiment with the flower-pot as clearing up our pre-existing ignorance; we should regard the fact of their producing cresses and grasses as proof positive that the particles sown in the earth of the pot were the seeds of the plants which have grown from them. It would be simply monstrous to conclude that they had been 'spontaneously generated.'

This reasoning applies word for word to the development of *Bacteria* from that floating matter which the electric beam reveals in the air, and in the absence of which no Bacterial life has been generated. I cannot see a flaw in the reasoning; and it is so simple as to render it unlikely that the notion of Bacterial life being developed from dead dust can ever gain currency among the members of the medical profession.

It has been said of those whom the evidence adduced in favour of spontaneous generation fails to convince, that they seem willing to believe in almost any infringement of natural uniformity rather than admit the doctrine.[1] This surely is an inversion of the true order of the facts. Natural uniformity is the record of experience; and, apart from the phenomena to be accounted for, there is not a vestige of experience, possessed either by the man of science or the human race, which warrants the notion that dead dust, and not living seed, is the source of the crops which spring from our infusions

[1] Transactions of the Pathological Society, vol. xxvi. p. 273.

when impregnated by the floating particles of the atmosphere.

§ 17. *Recent Experiments on Heterogenesis.*

The uniform sterility of the boiled infusions described in the foregoing pages, when protected from the floating matter of the air, proves that they do not contain germs capable of generating life. Our most advanced heterogenist, indeed, affirms that a temperature of 140° Fahr. reduces, in all cases, such germs to a state of actual or potential death; and he ingeniously argues that as, even in flasks which have been raised to a temperature of 212°, and hermetically sealed, putrefaction, and its associated *Bacteria*, do most certainly arise, such *Bacteria* must be spontaneously generated. 'We know,' he says, 'that boiled turnip or hay-infusions, exposed to ordinary air, exposed to filtered air, to calcined air, or shut off altogether from contact with air, are more or less prone to swarm with *Bacteria* and *Vibriones* in the course of from two to six days.'[1]

We are here met by a difficulty at the outset. The proof of Bacterial death at 140° Fahr. consists solely in the observed fact, that when *a certain liquid* is heated to that temperature no life appears in it afterwards; while *in another liquid* life appears two days after it has been heated to 212°. Instead of concluding that in the one liquid life is destroyed and in the other not, it is assumed that 140° Fahr. is the death-temperature for both; and this being so, the life observed in the second liquid is regarded as a case of spontaneous generation. A great deal of Dr. Bastian's most cogent reasoning rests upon this foundation. Assumptions of this kind guide him in his most serious experiments. He finds,

[1] 'Evolution and the Origin of Life,' p. 94.

for example, that a mineral solution does not develop *Bacteria* when exposed to the air; and he concludes from this that an organic infusion also may be thus exposed without danger of infection. He exposes turnip-juice accordingly, obtains a crop of *Bacteria*, which, in the light of his assumption, are spontaneously generated. Such are the warp and woof of some of the weightiest arguments on this question which have been addressed by him to the Royal Society.[1]

Granting, then, all that Dr. Bastian alleges regarding his experiments to be correct, the logical inference would be very different from his inference. In a future essay his position will be more clearly defined. To the examination of his experiments I now address myself.

§ 18. *Experiments with Filtered Air.*

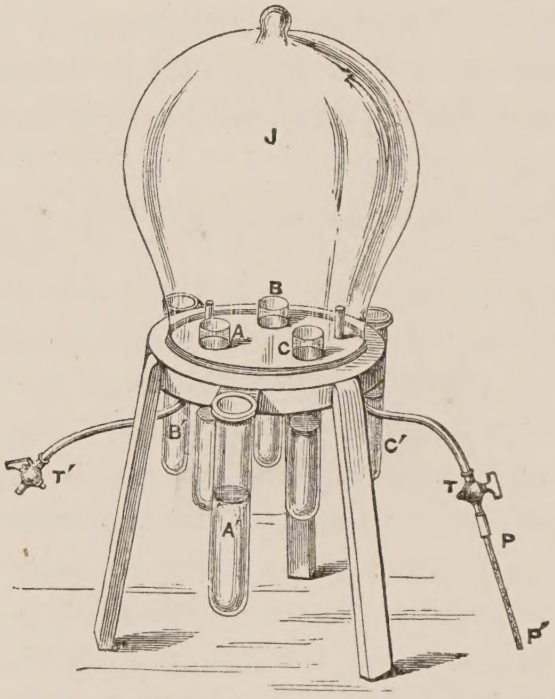
A bell-jar containing about 700 cubic inches of air was firmly cemented to a slab of wood coated with resin, and supported on three legs.[2] Through the slab passed, air-tight, three large test-tubes (A, B, C, fig. 4). Prior to cementing, the tubes had been three-fourths filled, one with an infusion of hay, another with an infusion of turnip, and a third with an infusion of mutton. On the 2nd of November the mote-laden air was pumped out, air slowly filtered through a long tight plug of cotton-wool being allowed to take its place. The jar was emptied and refilled until the closest scrutiny by a concentrated beam revealed no floating matter within it. The infusions were then boiled for five minutes,

[1] Proceedings, vol. xxi. p. 130.

[2] Two hoops of sheet iron, with an annular space about an inch wide, were fastened on to the slab of wood. The annular space was filled with hot cement, into which the hot bell-jar was pressed. The circular space within the smaller hoop was also covered by a layer of cement.

and abandoned to the air of the jar. During ebullition a small quantity of the liquid in one of the tubes boiled over, and rested upon the interior resinous surface at a little distance from the mouths of two of the tubes. The germinal matter, it may be remarked, is not readily blown away from such a surface, and it certainly was not wholly removed by our feeble current of filtered air. Three exposed tubes containing the same infusion were placed at the same time beside the protected ones.

FIG. 4.



In three days these exposed tubes became turbid and charged with life; *but for three weeks the infusions in contact with the filtered air remained perfectly clear.*

At the end of three weeks, that is on the 23rd of November, I desired my assistant to renew the air in the bell-jar. He pumped it out, and while permitting fresh air to enter through the cotton-wool filter, my attention

was directed to a couple of small round patches of *Penicillium* resting in the liquid that had boiled over on the resin. I at once made the remark that the experiment was a dangerous one, as the entering air would probably detach some of the spores of the *Penicillium* and diffuse them in the bell-jar. This was, therefore, filled very slowly, so as to render the disturbance a minimum. Next day, however, a tuft of mycelium was observed at the bottom of one of the three tubes, namely that containing the hay-infusion. It soon grew large enough to fill a considerable portion of the tube. For nearly a month longer the two tubes containing the turnip- and mutton-infusions maintained their transparency unimpaired. Late in December the mutton-infusion, which was in dangerous proximity to the outer mould, showed a tuft of *Penicillium* upon its surface. The beef-infusion continued bright and clear for nearly a fortnight longer. The cold winter weather caused me to add a third gas-stove to the two which had previously warmed the room where the experiments are conducted. The warmth of this stove played upon one side of the bell-jar; and on the day after the lighting of the stove, the beef-infusion gave birth to a tuft of mycelium. In this case the small spots of *Penicillium* might have readily escaped attention; and had they done so we should have had here three cases of 'spontaneous generation' more striking than most of those that have been adduced in support of this doctrine.

The experiment was subsequently made upon a larger scale. Twelve very large test-tubes were caused to pass air-tight through a slab of wood; the wood was thickly coated with cement, in which, while it was hot and soft, a heated 'propagating glass,' resembling a huge bell-jar, was imbedded. The air within the glass was pumped out several times, air filtered carefully through

a plug of cotton-wool being permitted to supply its place. The test-tubes contained infusions of hay, turnip, beef, and mutton, three of each, twelve in all. For two months they remained as clear and cloudless as they were upon the day of their introduction, while twelve similar tubes, prepared at the same time, in precisely the same way, and hung on to the slab of wood outside the propagating-glass, were, in less than a week, clogged with mycelium, mould, and *Bacteria*.

One of the protected tubes was accidentally broken, and though its aperture was rapidly plugged with cotton-wool, some common air must at the time have entered the propagating-glass. Evaporation from the infusions went on; the vapour was condensed by the glass above, trickled down its interior surface, carrying with it, in part, such matter as had attached itself to that surface. A kind of pool was thus formed upon the cement below. This after an interval of three months became spotted with disks of *Penicillium*, by the spores of which one or two of the infusions have been recently invaded, the production of very beautiful mycelium-tufts being the consequence.

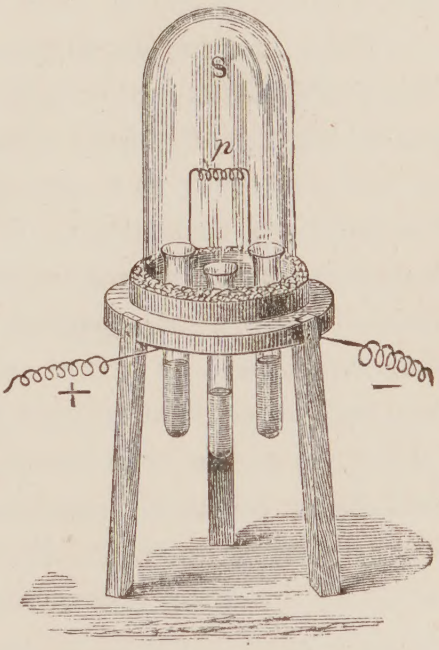
§ 19. *Experiments with Calcined Air.*

Six years ago[1] I showed that the floating matter of London air could be removed by permitting a platinum wire heated to whiteness to act upon it for a sufficient time. I availed myself of this mode of calcining the air on the present occasion. The apparatus employed is shown in fig. 5. A glass shade, s, is placed upon a slab of wood mounted on a tripod. Through the slab passes three large test-tubes nearly filled with the infusion to be examined. A platinum spiral, *p*, unites

[1] Proc. Roy. Inst. vol. vi. pp. 4 and 5.

the ends of two upright copper wires, which pass through the stand and are seen coiled outside it. The shade is surrounded below by a tin collar, with a space of about half an inch all round between it and the shade. This space is filled with cotton-wool firmly packed. Connecting the wires with a battery of fifteen cells, the spiral *p* was raised to whiteness, and was permitted to continue so for five minutes. Experiments previously executed had shown that this sufficed for the entire removal of the floating matter. When the spiral was heated, a portion of the expanded air was driven through the cotton-wool packing below; and when the current was interrupted, this air, returning into the shade, was prevented by the cotton-wool from carrying any floating matter with it.

Fig. 5.



The first three substances brought into contact with air calcined in this way were damson-juice, pear-juice, and infusion of yeast. They were boiled for five minutes, and *for five months they have remained without speck or turbidity.* Other tubes similarly boiled, and placed underneath shades containing the floating matter of the air, have long since fallen into mould and rottenness.

Turnip- and hay-infusions, rendered slightly alkaline, have been mentioned as particularly prone to spontaneous generation. I wished to test this. On the 26th of November, therefore, four shades were prepared,

two containing strong turnip-infusion and hay-infusion unneutralized, two containing the same infusions slightly superneutralized by caustic potash. The alleged spontaneous development of life was not observed. The tubes exhibit to this hour the clearness and colour which they showed on the day they were boiled. Hermetically-sealed tubes, containing the same infusions, prepared on the same day, remain equally clear; while the specimens exposed to the laboratory air have fallen into rottenness.

The experiments with calcined air were also executed in another form and on a larger scale. A 'propagating-glass,' similar to that already described, was cemented in the same way to a slab of wood through which passed twelve large test-tubes. The infusions, as before, were hay, turnip, beef, and mutton. The air being removed from the propagating-glass by a good air-pump, its place was supplied by other air which had passed slowly through a red-hot platinum tube containing a roll of platinum gauze, also heated to redness. Tested by a searching beam, this calcined air was found quite free from floating matter. For two months no speck invaded the limpidity of the infusions exposed to it, while a week's exposure to the ordinary air sufficed to reduce twelve similar infusions, hung on to the slab of wood outside the glass, to the muddiness of putrefaction.

§ 20. *Infusions withdrawn from Air.*

The arrangement here was the same as that adopted in the first experiment with filtered air, the only difference being that the bell-jar, with a view to its more perfect exhaustion, was smaller. It was cemented air-tight to a slab of wood through which passed three large test-tubes, filled to about two-thirds of their

capacity with infusions of beef, mutton, and turnip respectively. The bell-jar was exhausted six times in succession, and filled after each exhaustion with air carefully filtered through cotton-wool. While this air was in contact with the infusions they were boiled in a brine-bath. The receiver was afterwards exhausted as perfectly as a good air-pump could exhaust it; while outside the receiver were hung three tubes to compare with those within.

Here the protected infusions remained as clear as they were on the day of their introduction, not only after the exposed infusions had charged themselves with life, but for many weeks after they had evaporated away.

Such, then, are the tests to which I have subjected the statement that 'boiled turnip- and hay-infusions exposed to filtered air, to calcined air, or shut off altogether from contact with air, are more or less prone to swarm with *Bacteria* and *Vibriones* in the course of from two to six days.' These results, and others that might be adduced, leave no doubt upon my mind that the deportment of air from which the floating matter has been removed by filtration or calcination is precisely the same as that of air from which the particles have disappeared by self-subsidence. Once really sterilized, an infusion in contact with optically pure air, however obtained, remains sterile.

§ 21. *The Germ-theory of Contagious Disease.*

It is in connection with the germ-theory of contagious disease that the doctrine of spontaneous generation assumes its gravest aspect. My interest in the general question was first excited by the investigations of Pasteur, while the medical bearings of the doctrine

were subsequently made clear to me, mainly, I ought to say, by the writings and conversation of the late Dr. William Budd, who was the first of our countrymen to grasp with true philosophic insight the doctrine of 'the vitality of contagia,' which is now every day gaining ground.

At the present moment, indeed, no other medical principle occupies so much thought, or is the subject of so much discussion. 'How does it happen,' says Dr. Burdon Sanderson[1], 'that these *Bacteria*, which we suppose must have existed half a dozen years ago in as great numbers as at present, were then scarcely heard of, and that they now occupy so large a place in the medical literature of this country and of Germany, and have lately afforded material for lively discussion in the French Academy?' Dr. Sanderson points out the relation of Lister in England, and of Hallier in Germany, to the movement regarding *Bacteria* which is now working like a ferment through the medical world. But to no other workers in this field are we more indebted than to Dr. Sanderson himself, and to his colleagues, for the continued and successful prosecution of researches bearing upon the pathology of contagion.

'In 1870,' writes Mr. John Simon, in one of his excellent reports to the Privy Council, 'I had the honour of presenting Dr. Sanderson's first report of researches made in this matter. At that time general conclusions seemed justified, first, that the characteristic-shaped elements which the microscope had shown abounding in various infective products are self-multiplying organic forms, not congeneric with the animal body in which they are found, but apparently of the lowest vegetable kind; and, secondly, that such living organisms are probably the essence, or an inseparable part of the

[1] British Medical Journal, January 16, 1875.

essence, of all contagia of disease. . . . This view of the matter has since then become greatly more distinct, in consequence of the investigations made by Dr. Sanderson, particularly in 1871 and 1872, with reference to the common septic contagium or ferment. For in that ferment there seems now to be identified a force which, acting disintegratively upon organic matter, whether dead or living, can, on the one hand, initiate putrefaction of what is dead, and, on the other hand, initiate febrile and inflammatory processes in what is living.'

At a Meeting of the Pathological Society, held on the 6th of April, 1875, the germ-theory of disease was formally introduced as a subject for discussion, the debate being continued with great ability and earnestness at subsequent meetings. The Conference was attended by many distinguished medical men, some of whom were profoundly influenced by the arguments, and none of whom disputed the facts brought forward against the theory on that occasion. The leader of the debate, and the most prominent speaker, was Dr. Bastian, to whom also fell the task of replying on all the questions raised. The coexistence of *Bacteria* and contagious disease was admitted; but, instead of considering these organisms as 'probably the essence, or an inseparable part of the essence' of the contagium, Dr. Bastian contended that they were 'pathological products,' spontaneously generated in the body after it had been rendered diseased by the real contagium. The grouping of the ultimate particles of matter to form living organisms, Dr. Bastian considers to be an operation as little requiring the action of antecedent life as their grouping to form any of the 'other less complex chemical compounds.' Such a position must, of course, stand or fall by the evidence which its

supporter is able to produce; and accordingly Dr. Bastian appeals to the law and testimony of experiment as demonstrating the soundness of his view. He seems quite aware of the gravity of the matter in hand; this is his deliberate and almost solemn appeal:—'With the view of settling these questions, therefore, we may carefully prepare an infusion from some animal tissue, be it muscle, kidney, or liver; we may place it in a flask whose neck is drawn out and narrowed in the blow-pipe-flame, we may boil the fluid, seal the vessel during ebullition, and, keeping it in a warm place, may await the result, as I have often done. After a variable time the previously heated fluid within the hermetically sealed flask swarms more or less plentifully with *Bacteria* and allied organisms—even though the fluids have been so much degraded in quality by exposure to the temperature of 212° Fahr., and have thereby, in all probability, been rendered far less prone to engender independent living units than the unheated fluids in the tissues would be.'[1]

We have here, to use the words of Dr. Bastian, 'a question lying at the root of the pathology of the most important and most fatal class of diseases to which the human race is liable.' Let us now examine his settlement of the question, as described by himself in the foregoing extract.

§ 22. *Experiments with Hermetically-sealed Vessels.*

Experiments with hermetically-sealed tubes were begun by me on the 5th of October, 1875. The shape of the tubes after sealing is represented in fig. 6. Each of them contained about an ounce of liquid. They

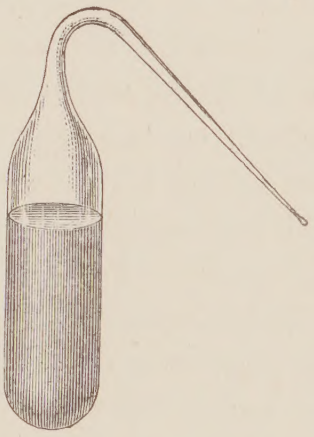
[1] Transactions of the Pathological Society of London, 1875, p. 272.

were boiled for only three minutes in an oil-bath, and were sealed, during ebullition, not by a blowpipe, but by the far more effectual spirit-lamp flame.

Hay.—Four tubes were charged on the date mentioned with a strong infusion, four with a weak infusion. All eight flasks remain to the present hour clear.

Turnip.—Two kinds of turnip were tried in these first experiments. Two tubes were charged with a strong infusion, and two with a weak infusion of a sound hard turnip; while two other pairs of tubes were filled with strong and weak infusions from a soft woolly turnip. All the tubes remain transparent to the present time. Two or three days' exposure to the air of the laboratory sufficed to cloud all these infusions and fill them with life.

Fig. 6.



On the 8th of October twenty-one tubes were charged with infusions of the following substances:—Mackerel, beef, eel, oyster, oatmeal, malt, potato. There were three tubes of each infusion. All of them remain to the present hour unchanged.

I had not previously seen a more beautiful illustration of the dichroitic action which produces the colours of the sky than in the case of the oyster-infusion. With reflected light it presented a beautiful cerulean hue, while it was yellow by transmitted light. This was due to the action of suspended particles which defied alike the power of the microscope and of ordinary filtration. At right angles to a transmitted beam the infusion copiously discharged perfectly polarized light.

Suspended particles in the potato-infusion produced a somewhat similar effect, but it was by no means so fine as that of the oyster-infusion. By ordinary filtration it was not possible completely to rid the malt and oatmeal infusions of suspended matter; but both remain exactly as they were when the flasks containing them were sealed.

These experiments had been made before the volume of the Transactions of the Pathological Society, containing the discussion referred to above came into my hands.[1] It caused me to turn again to my tubes, seeking further evidence. On the 12th of November thirty-six of them were charged, boiled, and hermetically sealed; on the 13th fifty-seven, on the 16th thirty-one, and on the 17th six tubes were similarly treated. The entire group of tubes, therefore, numbered one hundred and thirty. I tried moreover to multiply the chances of spontaneous generation by making the infusions of the most diverse materials. The following Table gives the names of the substances operated on, the number of tubes sealed, and the date of sealing:—

Fowl	6	tubes.	November 12th.
Mutton	6	,,	,,
Wild Duck	6	,,	,,
Beef	6	,,	,,
Herring	6	,,	,,
Haddock	6	,,	,,
Mullet	6	,,	November 13th.
Codfish	6	,,	,,
Pheasant	6	,,	,,
Rabbit	6	,,	,,
Hare	6	,,	,,
Snipe	6	,,	,,

[1] To the courtesy of Dr. Bastian I am indebted for a separate copy of the report of the discussion here referred to.

Partridge.	6	tubes	November 13th.
Plover	5	„	„
Liver	4	„	„
Heart	6	„	„
Tongue of Sheep . . .	6	„	November 16th.
Brains of Sheep . . .	3	„	„
Sweetbread	6	„	„
Humour of Ox-eye (undiluted)	2	„	„
Lens of Ox-eye . . .	3	„	„
Lungs of Sheep . . .	5	„	„
Tripe	6	„	„
Sole.	6	„	November 17th.

The tubes were immersed in groups of six at a time in an oil-bath, boiled for three minutes, and then sealed.

More than one hundred of these flasks were sensibly transparent and free from turbidity at the outset, and they remain so to the present hour. In some cases, however, it was not possible to wholly remove turbidity by filtration. I have already referred to the opalescence of oyster-infusion, which has invariably appeared whenever oyster has been digested. A still more pronounced case of the kind is furnished by an infusion of the crystalline lens of the ox. Nothing hitherto encountered by me imitates the flush of the true opal so closely as this infusion. Filtration through one hundred layers of paper was quite incompetent to remove the suspended particles to which this opalescence is due. Some of the other infusions remained turbid after filtration, without exhibiting what I should call opalescence. The sheep's lungs furnish an example of this. In some cases, moreover, where repeated filtering failed to remove the suspended particles, a few weeks' quiet caused them to sink, and leave the supernatant liquid clear. It may be worth remarking that some rabbit-infusions have shown a decided opalescence, while others have been

perfectly clear. The same remark applies to turnip-infusions, some of which have been found as clear as distilled water, while in general a slight opalescence is not to be got rid of by filtering.

These later experiments are quite in harmony with the earlier ones. Not a single flask of this multitude manifests the deportment alleged to be a matter of common observation.[1] If the power of spontaneous generation be a scientific verity, surely amid opportunities so multiplied and various it must have exerted itself. That the infusions employed were not 'degraded' by the boiling so as to be incapable of supporting life, was proved by the fact that exposed tubes containing the same infusions, treated in precisely the same way, resolved themselves with the usual speed into Bacterial swarms.

§ 23. *Conditions as to the Temperature and Strength of Infusions.*

In connection with these experiments, I have sought, to the best of my ability, to meet every condition and requirement laid down by others as essential to success. With regard to warmth, a temperature of 90° was generally attainable in our laboratory, while on certain days of mild weather without, and in favourable positions within, the temperature to which the infusions were subjected reached over 100° Fahr. As Dr. Bastian, however, had laid considerable stress on warmth, though most of his results were obtained with temperatures from 15° to 30° lower than mine,[2] I

[1] This group of flasks was submitted to the inspection of the Fellows of the Royal Society on the 13th of January, 1876.

[2] Proc. Roy. Soc. vol. xxi. p. 130. Also 'Beginnings of Life,' vol. i. p. 354.

thought it desirable to meet this new requirement also. The sealed tubes, which had proved barren in the Royal Institution, were suspended in boxes copiously perforated, so as to permit of the free circulation of warm air, and placed under the supervision of an intelligent assistant in the Turkish Bath in Jermyn Street. The washing-room of the establishment was found to be particularly suitable for our purpose; and here, accordingly, the boxes were suspended. From two to six days are allowed by Dr. Bastian for the generation of organisms in hermetically-sealed tubes. Mine remained in the washing-room for nine days. Thermometers placed in the boxes, and read off twice or three times a day, showed the temperature to vary from a minimum of 101° to a maximum of 112° Fahr. At the end of nine days the infusions were as clear as at the beginning.

They were then removed to a warmer position; the temperature 115° having been mentioned as favourable to spontaneous generation. For fourteen days my temperatures hovered about this point, falling once as low as 106°, reaching 116° on three occasions, 118° on one, and 119° on two. The result was quite the same as that recorded a moment ago. The higher temperatures proved perfectly incompetent to develop life.[1]

Fifty-six observations, including both the maximum and minimum thermometers, were taken while the tubes occupied their first position in the washing-room, and seventy-four while they occupied the second position. The whole record, carefully drawn out, is before me, but I trust the statement of the major and minor limits of temperature will suffice.

Dr. Bastian's demand for these high temperatures is, as already remarked, quite recent. Prior to my com-

[1] My thanks are due to the managers of the bath for their obliging kindness in this matter.

munication to the Royal Society on January 13, he had successfully worked with temperatures lower than those within my reach in Albemarle Street. There I followed his directions, adhered strictly to his prescriptions; but, taking care to boil and seal the liquids aright, his results refused to appear in my experiments. On learning this he raised an objection as to temperature, and made a new demand. With this I have complied; but his position is unimproved.

With regard to the question of concentration, I have already referred, in sections 3 and 16 of this memoir, to the great diversity in this particular presented by all my infusions, through their slow evaporation. But more than a general conformity to prescribed conditions was observed here also. The strength of an infusion is regarded as fixed by its specific gravity; and I have worked with infusions of precisely the same specific gravity as those employed by Dr. Bastian. This I was specially careful to do in relation to the experiments described and vouched for, I fear incautiously, by Dr. Burdon Sanderson in vol. vii. p. 180 of 'Nature.' It will there be seen that, though failure attended some of his efforts, Dr. Bastian did satisfy Dr. Sanderson that in boiled and hermetically-sealed flasks *Bacteria* sometimes appear in swarms. With purely liquid infusions I have failed to reproduce this result. Hay- and turnip-infusions, of accurately the same character and strength as those employed on the occasion referred to, were prepared, boiled in an oil-bath, carefully sealed up, and subjected to the proper temperatures. In multiplied experiments they remained uniformly sterile.[1]

[1] One hundred and twenty flasks, hermetically sealed, containing animal and vegetable infusions, some two, some three years old, are now beside me. They show no sign of Bacterial life.

§ 24. *Developmental Power of Infusions and Solutions: Air-germs contrasted with Water-germs.*

Wishing to make no experiment, whether with self-cleansed, filtered, or calcined air, or with infusions withdrawn from air by the air-pump, or contained in hermetically-sealed vessels, without exposing the same infusions to ordinary air, this comparison was instituted with the substances mentioned at pages 96 and 97. One hundred test-tubes, an inch wide and 3 inches deep, were divided into groups, each group being filled with the same infusion. The groups were sufficiently numerous to embrace all the substances mentioned in the Table referred to. Exposed to the uncleansed air, they were attacked with different degrees of rapidity and vigour; but in a few days all of them without exception became muddy and crowded with life. On the whole, the hare- and pheasant-infusions presented the greatest contrast. The tubes containing the former were far gone before those containing the latter were sensibly invaded. The putrescibility of pheasant, moreover, was exceeded by that of snipe, partridge, and plover. The sheep's heart examined was also slow to putrefy. A single illustration of this difference of developmental power may be given here.

On the 13th of November thirty tubes, containing infusions of partridge, pheasant, snipe, hare, sheep's heart, and codfish, five tubes being devoted to each, together with four tubes of plover, three of mullet, and three of liver, were exposed to the laboratory air. On the 15th, 16th, and 22nd the numbers taken possession of by *Bacteria* were as follows:—

	15th.	16th.	22nd.
Partridge	0	3	all
Pheasant	0	1	„
Snipe	2	3	„
Hare	2	4	„
Heart	0	1	„
Codfish	2	4	„
Plover.	1	2	„
Mullet	1	2	„
Liver	1	3	„

They had doubtless all given way some days before the 22nd, but I had not taken the precaution to look at them.

Thus, then, the first two days produced no visible change in the pheasant-infusion, while in two of the hare-tubes putrefaction had vigorously set in. Three days' exposure caused only one of the pheasant-tubes to yield; four of the hare-infusion had yielded in the same time. The difference between them was also illustrated by the mould upon their surfaces. Some days after their exposure four of the five pheasant-tubes were thickly covered with *Penicillium*, while the five hare-tubes, with one exception, which could hardly be considered such, had repelled that enemy, maintaining their *Bacteria* undisturbed.

Still the deportment of the hare-infusion may have been due, not to any specific difference between hare and pheasant, but to the circumstances preceding death. The researches of Dr. Brown-Séquard show that even the same animal tissue exhibits, under different circumstances, very different tendencies to putrefaction. In guinea-pigs subjected immediately after death to the action of the magneto-electric current, he found the rapidity of putrefaction to correspond with the violence of the tetanization. He also draws attention to the

influence of muscular exercise on cadaveric rigidity and putrefaction, showing how quickly they appear in 'overdriven cattle and in animals hunted to death.' It is known, indeed, to sportsmen that a shot hare will remain soft and limp for a day, while a hunted one becomes rigid in an hour or two. In September, 1851, two sheep which had been overdriven to reach a fair were killed by the section of the carotid arteries. 'Putrefaction,' says Dr. Brown-Séquard, 'was manifest before the end of the day, or in less than eight hours after death.'[1] The deportment of the hare operated upon by me may therefore depend upon the circumstance of its being brought down by the greyhound instead of the gun. It will be interesting to inquire how far the peculiarity of the animal tissue is transferred to the infusion. This is a fit subject for further investigation.[2]

Such observations inculcate caution in drawing inferences from the deportment of any infusion as to the distribution of germs in the air. The germs may be demonstrably present while the infusion may not favour their development. As to the quantity and quality of atmospheric germs, the hare and the pheasant might lead to different conclusions. A passing reference to an important practical inference may be fitly introduced here. In one of the earliest of the able series of researches with which he has enriched medical science, Dr. Burdon Sanderson exposed to the air 'Pasteur's solution,' which is capable of vigorously developing and nourishing *Bacteria*

[1] Croonian Lecture, Proc. Roy. Soc. 1862, vol. xi. p. 210.

[2] Five-and-twenty flasks containing pheasant-infusion were compared during the month of December with five-and-twenty containing infusion of hare. Neither in the rapidity of Bacterial development, nor in the readiness to support the growth of *Penicillium*, did the considerable differences between hare and pheasant first observed repeat themselves.

when they are communicated to it by inoculation; he also permitted air to bubble through the liquid, and finding no development in either case, he inferred the entire absence of *Bacteria* and their germs from the air, considering water to be their exclusive habitation. Other distinguished men have come to the same conclusion; while in his books and papers, and in the discussion before the Pathological Society already referred to, Dr. Bastian has forcibly dwelt upon the result as justifying the interpretation which he has affixed to his experiments. If, he rightly urges, the air be 'entirely free' from matter which could produce *Bacteria*, then their appearance in boiled infusions exposed to the air must be due, not to anything contained in the air, but to the inherent power of the infusions. Spontaneous generation is undoubtedly the logical outcome of the position that 'the germinal matter from which *Bacteria* spring does not exist in ordinary air.' The experiments, however, recorded in this memoir constitute an ocular demonstration of the respective parts played by the infusion and the air. A pinch of fungus-spores, taken between the fingers, sown in a suitable medium, and producing their appropriate crop, could not more clearly indicate the origin of that crop than experiments with the luminous beam indicate the origin of our harvests of *Bacteria*. Dr. Sanderson is, I doubt not, now well aware that his first statement was founded on an error of interpretation. In a lecture delivered at Owens College, Manchester, and published in the 'British Medical Journal' for January 16, 1875, he to a great extent qualifies and corrects his first inference. He there says that the *Bacteria* 'attach themselves without doubt to these minute particles, which, scarcely visible in ordinary light, appear as motes in the sunbeam, or in the beam of an electric lamp.' In fact the experiments on which

he based his first inference owed their barrenness, not to the absence of *Bacteria*-germs from the air, but to the inability or, rather, slowness of his mineral solution to develop them.

With regard to the part played by the visible motes, I may repeat here what has been previously stated, namely, that while the coarser particles could hardly exist in their midst without loading themselves to some extent with the minute germs of *Bacteria*, there is no reason to think the motes indispensable for the diffusion of the germs. Whether they are attached to each other or not, the dryness and the moisture of the air are shared equally by both. The germs, moreover, float in the air more readily than the larger particles; and they, I doubt not, when properly illuminated, shed forth a portion of that changeless light to which reference has been already made, and the perfect polarization of which declares the smallness of the masses which scatter it.

The prevalence of the germinal matter of *Bacteria* in water has been demonstrated by the experiments of Dr. Burdon Sanderson. But the germs in water, it ought to be remembered, are in a very different condition, as regards readiness for development, from those in air. In water they are already wetted, and ready, under the proper conditions, to pass rapidly into the finished organism. In air they are more or less desiccated, and require a period of preparation more or less long to bring them up to the starting-point of the water-germs.[1]

[1] The process by which an atmospheric germ is wetted would be an interesting subject of investigation. A dry microscope covering-glass may be caused to float on water for a year. A sewing-needle may be similarly kept floating, though its specific gravity is nearly eight times that of water. Were it not for some specific relation between the matter of the germ and that of the liquid into which it falls, wetting would be simply impossible. Antecedent to all development there must be an interchange of

The rapidity of development in an infusion infected by either a speck of liquid containing *Bacteria*, or by a drop of distilled water, is extraordinary. On the 4th of January I dipped a thread of glass almost as fine as a hair into a cloudy turnip-infusion, and introduced the tip only of the glass fibre into a large test-tube containing an infusion of red mullet: twelve hours subsequently the perfectly pellucid liquid was cloudy throughout. A second test-tube containing the same infusion was infected with a single drop of the distilled water furnished by Messrs. Hopkin and Williams; twelve hours also sufficed to cloud the infusion thus treated. Precisely the same experiments were made with herring infusion, with the same result. In the winter season several days' exposure to warmed air are needed to produce this effect with air-germs.

On the 31st of December a strong infusion was prepared by digesting turnip in distilled water at a temperature of 120° Fahr. It was divided between four large test-tubes, in one of which the infusion was left unboiled, in another boiled for five minutes, in the two remaining ones boiled and, after cooling, infected with one drop of beef-infusion containing *Bacteria*. In twenty-four hours the unboiled tube and the two infected ones were cloudy, the unboiled tube being the most turbid of the three. The infusion in the unboiled tube was peculiarly limpid after digestion; for turnip it was quite exceptional, and no amount of searching with the microscope could reveal in it at first the trace of a living *Bacterium*; still germs were there which, suitably nourished, passed in a single day into Bacterial swarms without number. Five days failed to produce an effect

matter between the germ and its environment; and this interchange must obviously depend upon the character of the encompassing liquid.

approximately equal to this in the uninfected boiled tube, which was exposed to the common laboratory air.

There cannot, I think, be a doubt that the germs in the air differ widely among themselves as regards *preparedness* for development. Some are fresh, others old; some are dry, others moist. Infected by such germs, the same infusion would require different lengths of time to develope Bacterial life. And this remark, I doubt not, applies to the different degrees of rapidity with which epidemic disease affects different people. In some the hatching-period, if I may call it such, is long, in some short, the differences depending upon the different degrees of preparedness of the contagium.[1]

§ 25. *Diffusion of Germs in the Air.*

During the earlier observations recorded in this essay, and others not here mentioned, about 100 exposed tubes or flasks had been distributed irregularly in the rooms where the inquiry is conducted. They expanded to nearly 1000 in the end: not one of them escaped infection. A few days always sufficed to cloud the exposed infusions, and fill them with Bacterial life. I placed tubes at various points in the Royal Institution—on the roof of the house outside, in my bedroom, in an upper kitchen, in my study, in the upper and lower libraries, in the theatre, model-room, reading-room, managers' room, and in the kitchen at the bottom of the house below the level of Albemarle Street. All were smitten with putrefaction, and with its invariable

[1] The medical student of the future will probably connect these remarks with the following statement of Dr. Murchison:—'In that protean disease typhoid fever, I have repeatedly had occasion to observe a remarkable similarity in the course, and even in the complications, according to the source of the poison.'—Trans. Path. Soc. vol. xxvi. p. 315.

associate, *Bacteria*. In the rooms without fires the action was slower than in the warmer rooms; but all the infusions gave way in the end.

In view of the statements which had been made regarding the scantiness of *Bacteria*-germs in the air, observations outside of London would, I thought, be interesting. Accordingly, on the 27th of October, a tube containing an infusion of beef was placed in the hands of Mr. Darwin, who had the kindness to set it in his study at Down and observe its changes. In three days it became cloudy and peopled with *Bacteria*. The same result was obtained in the open air. Mr. Francis Darwin was good enough to expose an infusion for me in his father's orchard: the weather was cold, and the progress, therefore, slow; but the tube which had been exposed on the 2nd of November was cloudy and full of *Bacteria* on the 9th. In Sir John Lubbock's study a similar result was obtained. From Sherwood, near Tunbridge Wells, infusions of fowl and wild duck were returned to me by Mr. Siemens thickly turbid and crowded with *Bacteria*. From Pembroke Lodge, Richmond Park, Mr. Russell returned tubes of turnip, beef, and mutton swarming with life. An infusion of beef exposed at Heathfield Park, Sussex, for a week was returned to me by Miss Hamilton muddy and filled with *Bacteria*. From Greenwich Hospital Mr. Hirst sent me tubes of beef-, mutton-, and turnip-infusion filled with vigorous *Bacteria*. Dr. Hooker was good enough to take charge of three sets of tubes at Kew, each set embracing beef, mutton, and turnip. One set was placed in the conservatory, with a temperature of 45° to 50°; one in his own study, with a temperature of 54° to 60°; a third set was placed in the orchid-house (the hottest in the gardens), with a temperature of 62° to 75°.

The tubes were exposed on the 4th of December, all of them being then clear. In the orchid-house the turnip became cloudy on the 7th, the beef and mutton on the 8th, after which the opacity rapidly increased. In the study all remained clear until the 9th, when the turnip began to cloud. On the 11th the beef was still clear, while the mutton had given way. On the 13th all of them had yielded. In the conservatory the turnip began to cloud on the 10th; the others followed in the same order as in the other cases.

The influence of temperature seems well shown by these observations. Three days sufficed to cloud the turnip in the orchid-house, five days in the study, and six days in the conservatory. The mutton in the study gathered over it a thick blanket of *Penicillium*. On the 13th it had assumed a light brown colour, 'as if by a faint admixture of clay;' but the infusion became transparent. The 'clay' here was the slime of dormant or dead *Bacteria*, the cause of their quiescence being the blanket of *Penicillium*. I found no active life in this tube, while all the others swarmed with *Bacteria*. From the Crystal Palace at Sydenham Mr. Price sent me tubes of mutton, beef, and turnip charged with *Bacteria*. The temperature was low at night, the development of life being thereby considerably retarded.

Thus, wherever it has been tested, the atmosphere has been found charged with the germs of *Bacteria*.

I wished, however, to obtain clearer and more definite insight as to the diffusion of atmospheric germs. Supposing a large shallow tray to be filled with a suitable organic infusion and exposed to the air. Into it the germs would drop; and could the resulting organisms be confined to the locality where the germs fell, we should have the floating life of the atmosphere *mapped*,

so to speak, in the infusion. But in such a tray the organisms would intermingle and thus mar the revelation of their distribution. Valuable information I thought might be gained by breaking up the infusion into isolated conterminous patches, and exposing them to the air.

A square wooden tray was accordingly pierced with one hundred circular apertures; into each of which was dropped a test-tube 3 inches long and 1 inch wide, with its rim resting in each case upon the rim of the aperture. There were ten rows of tubes, with ten tubes in each row. On the 23rd of October, 1875, thirty of these tubes were filled with an infusion of hay, thirty-five with an infusion of turnip, and thirty-five with an infusion of beef. The tubes with their infusions had been previously boiled ten at a time in an oil-bath.

One hundred circles were marked upon paper so as to form a plan of the tray, and every day the state of each tube was registered upon the corresponding circle. Seven such maps or records were executed.

I will use the term 'cloudy' to denote the early stage of turbidity, distinct but not strong. The term 'muddy' will be used to denote thick turbidity.

§ 26. *Tray of one hundred Tubes.*

On the 25th of October one or two of the tubes exposed on the 23rd showed signs of yielding; but the progress of putrefaction was first registered on the 26th. Fig. 7, embracing the first record, is annexed; it may be thus described.

Hay.—Of the thirty specimens exposed, one had become 'muddy'—the seventh in the middle row, reckoning from the side of the tray nearest a stove. Six tubes remained perfectly clear between this muddy one and the stove, proving that differences of warmth

may be overridden by other causes. Every one of the other tubes containing the hay-infusion showed spots of mould upon the clear liquid.

Turnip.—Four of the thirty-five tubes were very muddy, two of them being in the row next the stove, one four rows distant, and the remaining one nine rows away. Besides these, seven tubes had become clouded. There was no mould on any of the tubes.

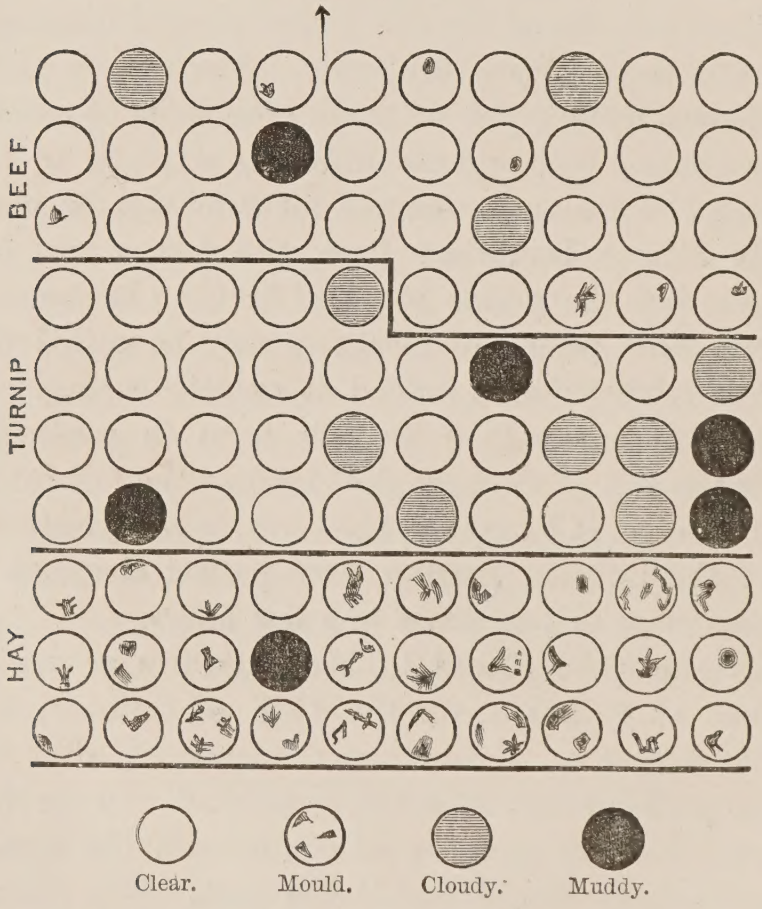
Beef.—One tube of the thirty-five was quite muddy, in the seventh row from the stove. There were three cloudy tubes, while seven of them bore spots of mould.

As a general rule organic infusions exposed to the air during the autumn remained for two days or more perfectly clear. Doubtless from the first germs fell into them, but the germs required time to become organisms. This period of clearness may be called the 'period of latency;' and, indeed, it exactly corresponds with what is understood by this term in medicine. Towards the end of the period of latency the fall into a state of disease, if I may use the term, is comparatively sudden; the infusion passing from perfect clearness to cloudiness more or less dense in a few hours.

Thus the tube placed in Mr. Darwin's possession was clear at 8.33 A.M. on the 19th of October, and cloudy at 4.30 P.M. Seven hours, moreover, after the first record of our tray of tubes, a marked change had occurred. For the purpose of comparison the second record, fig. 8, is placed beside the first. The change may be thus described:—Instead of one, eight of the tubes containing hay-infusion had fallen into uniform muddiness. Nineteen of them had produced Bacterial slime, which had fallen to the bottom, every tube containing the slime being covered by mould. Three tubes only remained clear, but with mould upon their surfaces. The muddy turnip-tubes had increased from four to ten; seven tubes were clouded, while eighteen of them

remained clear, with here and there a speck of mould on the surface. Of the beef, six were cloudy and one thickly muddy, while spots of mould had formed on the majority of the remaining tubes. Fifteen hours subsequent to this observation, viz. on the morning of the 27th of October, all the tubes containing hay-infusion were smitten, though in different degrees, some of them being much

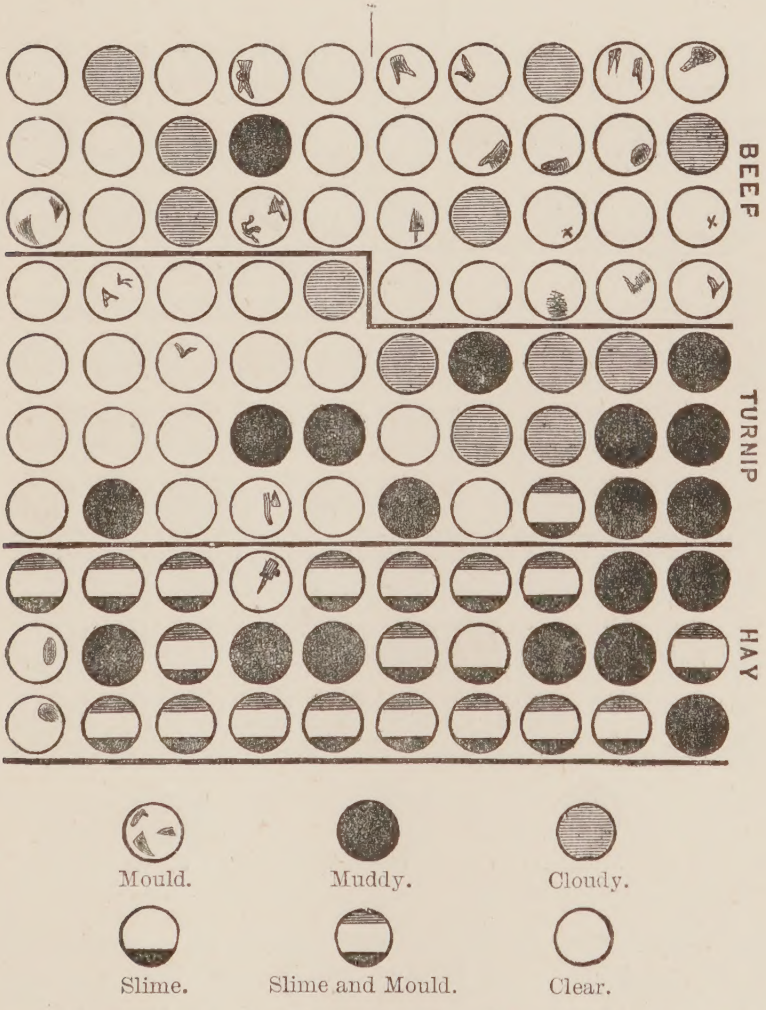
FIG. 7. 26th Oct., noon.



more turbid than others. Of the turnip-tubes, three only remained unsmitten, and two of these had mould upon their surfaces. Only one of the thirty-five beef-infusions remained intact. A change of occupancy, moreover, had occurred in the tube which first gave way. Its muddiness remained grey for a day and a half, then it changed to bright yellow-green, and

maintained this colour to the end. On the evening of the 27th every tube of the hundred was smitten, the majority with uniform turbidity, some, however, with mould above and slime below, the intermediate liquid being clear. The whole process bore a striking resemblance to the propagation of a plague among a population, the attacks being successive and of different

FIG. 8. 26th Oct., 7 P.M.



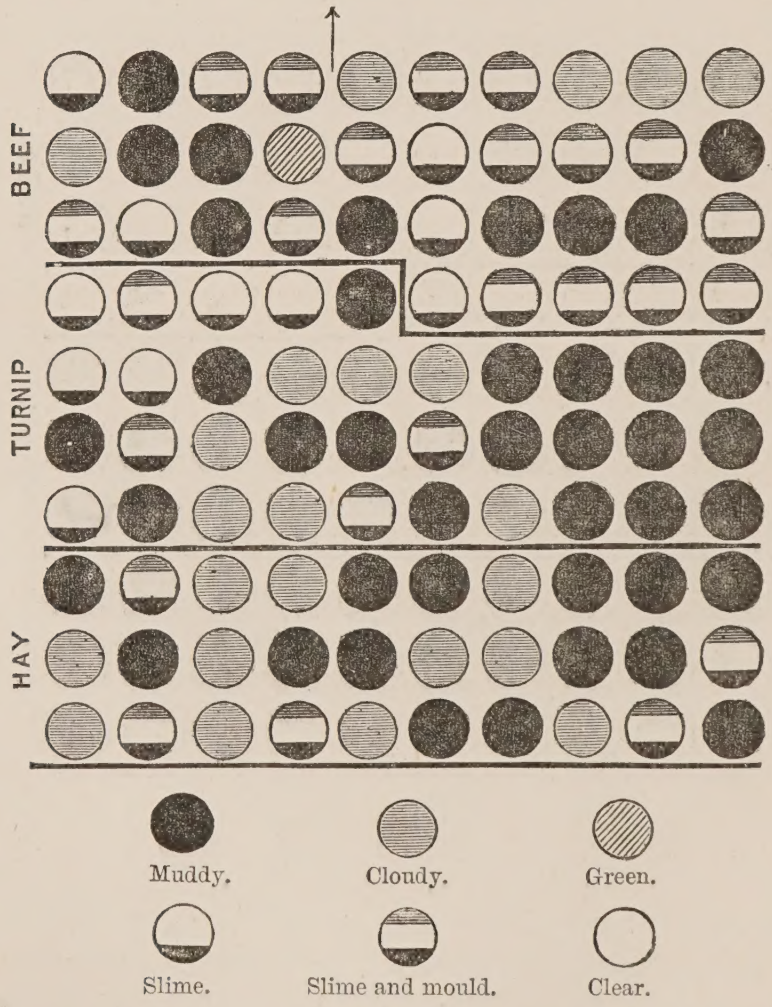
degrees of virulence. I annex copies of the fourth and seventh maps (figs. 9, 10) with their respective dates.

On the 31st of October I finally inspected the tray of tubes. All those containing the hay-infusion were turbid, some thicker and much more deeply coloured

than others. They had been all at first alike in colour. Out of the thirty tubes four only were free from mould. Three of these were adjacent to each other, the fourth at a distant portion of the tray.

The *Penicillium* was exquisitely beautiful. Its prevalent form was a circular patch made up of alternate zones of light and deep green. In some cases the liquid

FIG. 9. 27th Oct., 6.30 P.M.

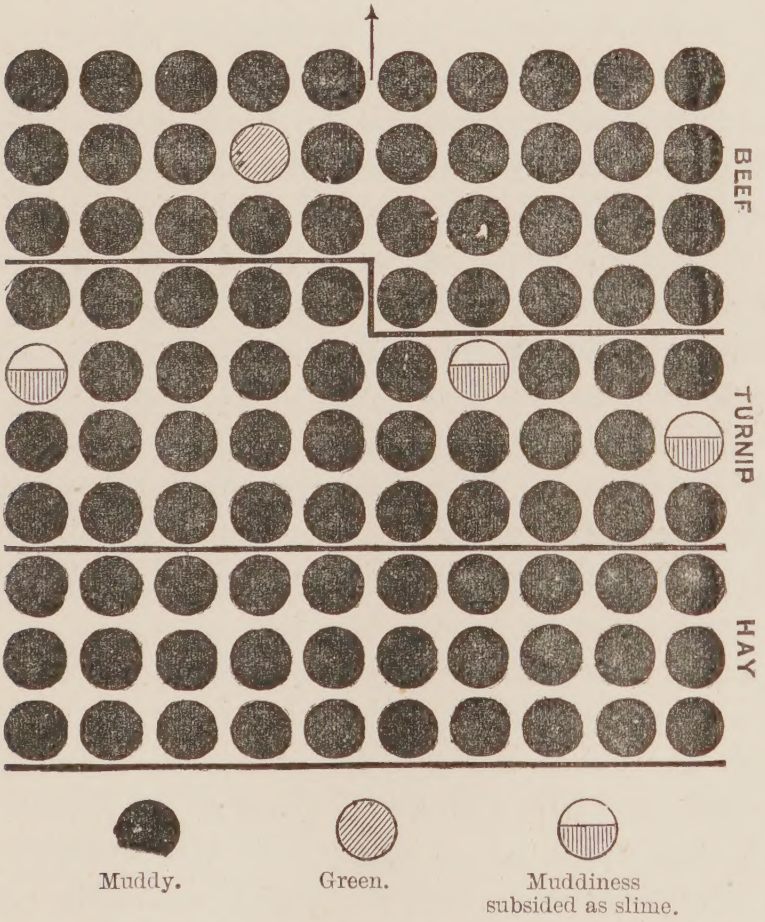


was covered by a single large patch; in others there were three or four patches, each made up of its differently coloured zones. Reticulated patterns also occurred. Three kinds of *Penicillium* seemed struggling for existence, namely:—that just described; a second kind, of the same consistency and colour, but

forming little rounded heaps instead of circles; thirdly, a woolly, voluminous, white mould, in the middle of which a zoned circle of the other mould sometimes formed a little islet.

All the tubes containing the turnip-infusion were also turbid on the 31st. Nine of them were free from mould. This, where it occurred, exactly resembled

FIG. 10. 29th Oct., 10.30 A.M.



small cocoons in shape. The beef-tubes were also all turbid on the 31st, and seventeen of them were free from mould. The mould upon the beef, moreover, was much less luxuriant than that on the hay- and turnip-infusions. The mould-developing power is obviously greatest in the hay-, less in the turnip-, and least of all in the beef-infusion. In every case where the mould

was thick and coherent the *Bacteria* died, or became dormant, and fell to the bottom as a sediment. The growth of mould and its effect on the *Bacteria* are very capricious. The turnip-infusion, after developing in the first instance its myriadfold Bacterial life, frequently contracts mould, which stifles the *Bacteria* and clears the liquid all the way between the sediment and the scum. Of two tubes placed beside each other, one will be taken possession of by *Bacteria*, which successfully fight the mould and keep the surface perfectly clean; while another will allow the mould a footing, the apparent destruction of the *Bacteria* being the consequence. This I have proved to be the case with all infusions, fish, flesh, fowl, and vegetable. At the present moment, for example, of three tubes containing an infusion of sole, placed close together in a row, the two outside ones are covered by a thick tough blanket of mould, while the central one has not a single speck upon its surface. The *Bacteria* which manufacture a green pigment appear to be uniformly victorious in their fight with the *Penicillium*.

These observations enable us, I think, to draw some interesting conclusions. From the irregular manner in which the tubes are attacked we may infer that, as regards *quantity*, the distribution of the germs in the air is not uniform. A single tube will sometimes be a day or more in advance of its neighbours. The singling out, moreover, of one tube of the hundred by the particular *Bacteria* that develop a green pigment, and other cases just adverted to, shows that, as regards *quality*, the distribution is not uniform. This has been further illustrated by the following observations:—Of five-and-twenty tubes of different animal infusions exposed in groups of five, in the middle of November, and

all swarming with Bacterial life, five were green. They were distributed as follows:—Beef 2, herring 1, haddock 1, fowl 1, wild duck 0. The same absence of uniformity was manifested in the struggle for existence between the *Bacteria* and the *Penicillium*. In some tubes the former were triumphant; in other tubes of the same infusion the latter was triumphant. It would also seem that a want of uniformity as regards *vital vigour* prevailed. With the self-same infusion the motions of the *Bacteria* in some tubes were exceedingly languid; while in other tubes the motions resembled a rain of projectiles, being so rapid and violent as to be followed with difficulty by the eye. Reflecting on the whole of this, I conclude that the germs float through the atmosphere in groups or clouds, and that now and then a cloud specifically different from the prevalent ones is wafted through the air. The touching of a nutritive fluid by a Bacterial cloud would naturally have a different effect from the touching of it by the sterile air between two clouds. But, as in the case of a mottled sky, the various portions of the landscape are successively visited by shade, so, in the long run, are the various tubes of our tray touched by the Bacterial clouds, the final fertilization or infection of them all being the consequence.[1]

[1] In hospital practice the opening of a wound during the passage of a Bacterial cloud would have an effect very different from the opening of it in the interspace between two clouds. Certain caprices in the behaviour of dressed wounds may possibly be accounted for in this way.

Under the heading 'Nothing New under the Sun,' Professor Huxley has lately sent me the following remarkable extract:—'Uebrigens kann man sich die in der Atmosphäre schwimmenden Thierchen wie Wolken denken, mit denen ganz leere Luftmassen, ja ganze Tage völlig reinen Luftverhältnisse wechseln.' (Ehrenberg, 'Infusionsthierchen,' 1838, p. 525.) The coincidence of phraseology is surprising, for I knew nothing of Ehrenberg's conception. My 'clouds,' however, are but small miniatures of his.

The tray of tubes proved so helpful in enabling me to realize mentally the distribution of germs in the air, that on the 9th of November, 1875, I exposed a second tray containing one hundred tubes filled with an infusion of mutton. On the morning of the 11th six of the ten nearest the stove had given way to putrefaction; three of the row most distant from the stove had yielded, while here and there over the tray particular tubes were singled out and smitten by the infection. Of the whole tray of one hundred tubes, twenty-seven were either muddy or cloudy on the 11th. Thus, doubtless, in an infective atmosphere, are individuals successively struck down. On the 12th all the tubes had given way, but the differences in their contents were extraordinary. All of them contained *Bacteria*, some few, others in swarms. In some tubes they were slow and sickly in their motions, in some apparently dead, while in others they darted about with rampant vigour. These differences are to be referred to differences in the germinal matter, for the same infusion was presented everywhere to the air. Here also I imagine we have a picture of what occurs during an epidemic, the difference in number and energy of the Bacterial swarms resembling the varying intensity of the disease. It becomes obvious from these experiments that of two individuals of the same population exposed to a contagious atmosphere, the one may be severely, the other lightly attacked, though, as regards susceptibility, the two individuals may be as identical as two samples of one and the same mutton-infusion. What I have already said regarding the 'preparedness' of contagium has its application here.

The parallelism of these actions with the progress of infectious disease may be traced still further. The 'Times,' for example, of January 17, 1876, contained a letter on typhoid fever, signed 'M.D.,' in which occurs

the following remarkable statement :—'In one part of it [Edinburgh], congregated together and inhabited by the lowest of the population, there are, according to the Corporation return for 1874, no less than 14,319 houses or dwellings—many under one roof, on the "flat" system—in which there are no house connexions whatever with the street-sewers, and, consequently, no water-closets. To this day, therefore, all the excrementitious and other refuse of the inhabitants is collected in pails or pans, and remains in their midst, generally in a partitioned-off corner of the living-room, until the next day, when it is taken down to the streets and emptied into the Corporation carts. Drunken and vicious though the population be, herded together like sheep, and with the filth collected and kept for 24 hours in their very midst, it is a remarkable fact that typhoid fever and diphtheria are simply unknown in these wretched hovels.'

The analogy of this result with the behaviour of our infusions is perfect. On the 30th of last November, for example, a quantity of animal refuse, embracing beef, fish, rabbit, hare, was placed in two large test-tubes opening into a protecting-chamber containing six tubes. On December 13, when the refuse was in a state of noisome putrefaction, infusions of whiting, turnip, beef, and mutton were placed in the other four tubes. They were then boiled and abandoned to the action of the foul 'sewer-gas' emitted by their two putrid companions. On Christmas-day, 1875, these four infusions were limpid. The end of the pipette was then dipped into one of the putrid tubes, and a quantity of matter, comparable in smallness to the pock-lymph held on the point of a lancet, was transferred to the turnip. Its clearness was not sensibly affected at the time; but on the 26th it was turbid throughout. On the 27th a speck from the infected turnip was transferred to the whiting; on the

28th disease had taken entire possession of the whiting. To the present hour the beef- and mutton-tubes remain as limpid as distilled water. Just as in the case of the living men and women in Edinburgh, no amount of fetid gas had the power of propagating the plague, as long as the organisms which constitute the true contagium did not gain access to the infusions.

In the foregoing observations the tubes were arranged in the same horizontal plane; but I also sought to obtain some notion of the vertical distribution of the germs in the air of the room. Two trays, each containing 100 tubes, were supported the one above the other in the same frame. The upper tray had all the air between it and the ceiling, a height of about 12 feet, from which the germs might descend upon it; the lower tray was shaded by the upper, a space of only 6 inches existing between them. If the number of germs deposited in the tubes were determined by the air-space above, the upper tray would be the one most rapidly and thoroughly taken possession of. The reverse was the case. As regards the development of Bacterial life, the lower tray was from first to last in advance of its neighbour. It is not air-space, then, so much as stillness, that determines the deposition of the germs. The air between the two trays being less disturbed than the general air of the room, the germs were less wafted about, and therefore sank in greater numbers into the tubes of the lower tray. We have here data which will enable us to form a rough notion of the lower limit of the number of germs contained in the room where the experiments were made.

The floor of the room measured 20 feet by 15 feet; its area was therefore 43,200 square inches, and every square inch would afford room for the section of one of our test-tubes. The height of the room is 180 inches;

hence 30 layers of tubes 6 inches apart might be placed one above the other between the floor and ceiling. This would make 1,296,000 tubes. If only a single germ a day fell into each tube, this would be the number of the germs. If the number deposited were one an hour, we should have thirty millions a day sown in the tubes. Probably the average time necessary for infection is very much less than an hour. At all events, 30,000,000 of germs daily would be an exceedingly moderate estimate of the number falling into our thirty layers of tubes. This, moreover, would only be a fraction—probably a small fraction—of the germs really present in the air. In his Presidential Address to the British Association at Liverpool, Professor Huxley ventured the statement that myriads of germs are floating in our atmosphere. Certain experimenters have rashly ridiculed this statement. In view of the foregoing calculation it, however, expresses the soberest fact. Indeed, taking the word myriad in its literal sense of ten thousand, it would be simple bathos to apply it to the multitudinous germs of our air.

§ 27. *Some Experiments of* Pasteur *and their Relation to Bacterial Clouds.*

Quite recently I had occasion to refresh my memory of Pasteur's paper published in the 'Annales de Chimie' for 1862. The pleasure I experienced on first reading it was revived by its reperusal. Clearness, strength, and caution, with consummate experimental skill for their minister, were rarely more strikingly displayed than in this imperishable essay. Hence it is that during recent discussions, in which this and other labours of the highest rank met with such scant respect, those in England most competent to judge of the value of scien-

tific work never lost faith in the substantial accuracy of Pasteur. One striking example of his penetration has an immediate bearing on the conclusion regarding Bacterial clouds, independently drawn by me from the deportment of the tray of one hundred tubes. On the 28th of May, 1860, Pasteur opened, on an uncovered terrace a few metres above the ground, four flasks containing the water of yeast. Nothing appeared in any of them until the 5th of June, when a small tuft of mycelium was observed in one of them. On the 6th a second tuft appeared in another flask ; the two remaining flasks continued intact and without organisms. On the 20th of July he opened, in his own laboratory, six flasks containing water of yeast. Four of them remained perfectly intact, while two of them became promptly charged with organisms. From these observations Pasteur inferred the non-continuity of the cause to which so-called spontaneous generation is due. This inference is quite in accord with the notion of Bacterial clouds suggested by my observations. Pasteur, in fact, sometimes opened his flask in the midst of a Bacterial cloud and obtained life, sometimes in the interspace between two clouds, and obtained no life.

Not with a view of repeating this observation, which I had forgotten, but for another reason, I opened on the 6th of January last a number of hermetically-sealed tubes in one and the same room of the Royal Institution. The names of the infusions contained in the tubes, the date of sealing them up, their condition before opening on the 6th, and their appearance six days subsequently are given in the accompanying table. I chose for these observations tubes which contained a little liquid in their drawn-out portions. In every case the motion of this liquid, when the tube was broken, indicated a violent inrush of air.

Infusion.	Date of sealing.	Appearance, Jan. 6.	Appearance, Jan. 12.
Grouse	Nov. 27th	Clear	Clear.
Sole	,, 17th	,,	Turbid.
Turnip No. 1	Oct. 5th	,,	Penicillium on surface.
Turnip No. 2	,, ,,	,,	Clear.
Hay	,, ,,	,,	Mycelium at bottom.
Wild Duck	Nov. 12th	,,	Turbid.
Mutton	,, ,,	,,	Cloudy.
Fowl	,, ,,	,,	Clear.
Beef	,, ,,	,,	Mycelium at bottom.
Haddock	,, ,,	,,	Clear.
Sweetbread	,, 16th	,,	Mycelium at bottom.
Rabbit	,, 13th	,,	Clear.
Heart	,, ,,	,,	Curdy layer at top.
Pheasant	,, ,,	,,	Clear.
Mullet	,, ,,	,,	,,
Hare	,, ,,	,,	,,
Snipe	,, ,,	,,	,,
Partridge	,, ,,	,,	,,
Plover	,, ,,	,,	Mycelium below.
Codfish	,, ,,	,,	Clear.
Kidney	Jan. 5th	,,	Mycelium at bottom.
Salmon	Dec. 13th	,,	Clear.
Whiting	,, ,,	,,	,,
Turnip	,, 29th	,,	,,
Hay, 4 drops of caustic potash	Nov. 22nd	Clear with sediment	Mycelium at bottom.
Hay, 2 drops of caustic potash	,, ,,	Clear	Mycelium at bottom.
Hay, 5 drops of caustic potash	,, ,,	Clear with sediment	Clear.
Hay, 6 drops of caustic potash	,, ,,	Clear with sediment	,,
Liver	Nov. 30th	Clear	,,
Hay	,, 18th	,,	,,
Hay	,, ,,	,,	,,
Turnip	,, ,,	,,	Muddy.

Thus, out of 31 flasks opened in the same air, 18 remained intact, while 13 were taken possession of by organisms—a fact obviously the same in character as that described by Pasteur. Such experiments demonstrate, if demonstration were needed, that it is not the air itself, or any gaseous or vaporous substance uniformly diffused through it, but some discontinuous substance floating in it, that is the cause of the infection. Instead of our tubes, let us suppose thirty-one wounds to be opened in the same ward of a hospital; plainly what has occurred with the tubes may occur with these wounds—some may receive the germs and putrefy, others may escape. Helped by the conception not only of germs, but of germ-clouds, the different behaviour of wounds subjected apparently to precisely the same conditions will cease to be an inscrutable mystery to the surgeon.[1]

During the course of this inquiry some eminent biologists have been good enough, from time to time, to look in upon my work, and to give me their views

[1] 'We have ample facts of experiment in our hands,' said Mr. Knowsley Thornton (Trans. of the Pathological Society, vol. xxvi. p. 313), 'to show that it is not the ga of the air, or any soluble material in water, but something "particulate" which sets up all the train of changes in an open wound, which may, after the patient has passed through a period of more or less constitutional disturbance, end in the healing of the wound, or may end in septicæmia and death. This particulate material, then, I believe we have evidence enough to prove consists of germs of *Bacteria* and other low organisms.' All the evidence points to this conclusion. I may say that I entirely agree with Mr. Thornton in the distinction he draws between *germs* and developed *Bacteria* floating in the air. It is, in my opinion, of the very last importance to seize this distinction with clearness. When it is fully realized we shall probably hear less of the arguments against Bacterial contagia founded on the fact that a virus diminishes in strength as the *Bacteria* multiply. A portion of the energy of the virus consists in its passage from the germ state to that of the finished organism.

regarding the evidential force of the experiments. To Professor Huxley, moreover, I am indebted for undertaking the examination of a number of the hermetically-sealed tubes. Thirty of them were placed in his hands, none of them being regarded as defective. A close examination, however, disclosed in one of them a mycelium. No faultiness could for a time be discovered in the tube; the sealing appeared to be quite as perfect as that of its sterile fellows. Once, however, on shaking it a minute drop of liquid struck my friend's face; and he soon discovered that an orifice of almost microscopic minuteness had been left open in the nozzle of the tube. Through this the common air had been sucked in as the liquid cooled, and hence the contamination. It was the only defective tube of the group of thirty, and it alone showed signs of life.

The statement of this fact before the Royal Society, by Professor Huxley, brought to my mind a somewhat similar experience of my own. One morning in November I lifted one of the hermetically-sealed tubes from the wire on which it was suspended, and, holding it up against the light, discovered, to my astonishment, a beautiful mycelium at the bottom. Before restoring the tube to its place I touched its fused end and found it cutting sharp. Close inspection showed that the nozzle had been broken off; the common air had entered, and the seed of the mycelium had been sown. Two other instances, one like that observed by Professor Huxley, have since come to light. In one of them a minute orifice remained after the supposed sealing of the tube. The other case was noticed when the tubes were returned from the Turkish bath. One of them contained a luxuriant mycelium. It was noticed that the liquid in this tube had singularly diminished in quantity, and on turning the tube up it was found cracked at the bottom.

No case of pseudo-spontaneous generation ever occurred under my hands that was not to be accounted for in an equally satisfactory manner.

In this inquiry, thus far, I have confined my observations to purely liquid infusions, purposely excluding milk, mixtures of turnip-juice and cheese, and, indeed, mixtures of solids and liquids of all kinds. The next section of the investigation will be devoted to these and kindred subjects; and to it I also postpone the complete examination of *pepton*, and of the remarkable experiments described by Dr. William Roberts, a small residue of which only I have failed to corroborate.

Throughout the whole of this investigation I have had to congratulate myself on the zealous and efficient aid of my excellent assistant, Mr. John Cottrell. His intelligence in seizing my ideas, and his mechanical skill in realizing them, have rendered me admirable service. Without such aid, indeed, so much ground could not have been covered in the time.

Royal Institution, 5th April, 1876.

It gives me special pleasure to direct attention here to a paper by the Rev. W. H. Dallinger, for an advanced proof of which I am indebted to the courtesy of Dr. Lawson, editor of the 'Popular Science Review.' Mr. Dallinger and his colleague Dr. Drysdale are known to have pushed the microscope to its utmost power of performance at the present time. Their 'Researches into the Life-History of the Monads' are models of scientific thoroughness and concentration. Mr. Dallinger's review of the present position of the doctrine of spontaneous generation, his remarks on Bacterial germs in relation

to the limits of the powers of the microscope, his demonstration that the germs of monads survive in a medium raised to a temperature which destroys the adult, and that precipitated mastic particles like those mentioned in § 16 of this paper are not to be discerned by a magnifying-power of 15,000 diameters, constitute a most interesting and important communication.

NOTE I. *Action of Bacteria upon a Beam of Light.*

To trace the gradual growth and multiplication of the *Bacteria* by their action on a beam of light, an infusion of beef was prepared on the 5th of October, 1875, placed in a globular flask of about 50 cubic inches capacity, and put aside with its mouth open to the laboratory air. On the 8th, 9th, 10th, 11th, and 12th similar flasks were prepared and put aside in succession. On the 12th all the flasks were examined by the concentrated electric light. The freshest one showed the track of the beam as a richly-coloured green cone. The green light was unaffected by a Nicol's prism, which, however, quenched the ordinary scattered light and augmented the purity and vividness of the green. It was a case of fluorescence. In the second flask, one day old, the fluorescent beam was in great part masked by the scattered light; the latter, however, could be partially quenched by a Nicol's prism, the purity of the fluorescence being thus in part restored. Through the third flask, two days, and through the fourth flask, three days old, the track of the beam was still discernible; through the fifth flask, four days old, it was all but obliterated, while in the sixth flask, seven days old, it was entirely shattered, the turbid medium being filled uniformly with the laterally scattered light.

Two of these flasks were of a bright yellow-green

colour, two were milky or white, and two of a dull brownish hue.

Cohn mentions the bluish tinge of the infusion by reflected and its yellow tinge by transmitted light when the *Bacteria* are incipient. This is due to a dichroitic action, similar to that which produces the blue of the sky and the morning and evening red. The blue, however, though discernible, is not pronounced, for the *Bacteria* are too large to scatter the colour in any high degree of purity; but with a 'muddy' infusion a very fair red may be obtained from transmitted light. I have used the Bacterial turbidity for photometric purposes. On the 9th of October, for example, I accompanied Sir Richard Collinson and a Committee of the Elder Brethren of the Trinity House to Charlton, with the view of comparing together two lights mounted at the Trinity Wharf at Blackwall. To imitate a foggy atmosphere, I employed an infusion cloudy with *Bacteria* and placed in a glass cell. With it the beams could be toned gradually down to complete extinction.

Note II. *Fluorescence of Infusions.*

All the animal infusions, both flesh and fish, showed the same fluorescence. It was the same green hue throughout, though of varying degrees of intensity. In wild duck, grouse, snipe, hare, partridge, and pheasant the fluorescence was fine—sometimes exceedingly fine. In rabbit it was less fine than in hare, and in a tame rabbit less fine than in a warren rabbit. Fishes also differed from each other. Mullet, for example, was finer than cod, herring, or haddock. Beef, mutton, heart, liver, all showed the same green fluorescence.

Led up to it by a series of remarkable experiments

on the rapidity of the passage of crystallized substances into the vascular and non-vascular textures of the body,[1] Dr. Bence Jones and Dr. Dupré communicated to the Royal Society in 1867 a highly interesting paper 'On a Fluorescent Substance, resembling Quinine, in Animals.'[2] They then showed that 'from every texture of man and of some animals a fluorescent substance can be extracted, which, when extracted, has a very close optical and chemical resemblance to quinine.' They therefore called it animal quinoidine. In dilute solutions they found that the fluorescence of the animal substance was not to be distinguished from that produced by quinine. When the solution was concentrated, the colour of the light was of a decidedly greenish hue. This latter observation is most in agreement with mine. In all the infusions examined by me the fluorescent light was a decided green, and not to be mistaken for the blue light of quinine.

The green colour is similar to that emitted by the crystalline lens when a beam of violet light impinges on it;[3] sending such a beam through any of the infusions, the 'degradation' of the violet to green is strikingly illustrated.

The foregoing statement refers to the deportment of the infusions after boiling and filtering. Prior to boiling some of them were of a brilliant ruby colour; but even here, when the layer of liquid between the eye and the beam was not too thick, the green fluorescence could be seen through the red liquid.

[1] Proceedings of the Royal Society, vol. xiv. 1865.

[2] *Ibid.* vol. xv. p. 73.

[3] On plunging the eye into the beam of the electric lamp, transmitted through violet glass, the moment the crystalline lens is seen to fluoresce by a second observer, a blue shimmer is seen by the eye on which the beam falls. In the case of my own eye, I can always readily tell when the fluoresence has set in.

FURTHER RESEARCHES ON THE DEPORTMENT AND VITALITY OF PUTREFACTIVE ORGANISMS.[1]

III.

§ 1. *Introduction.*

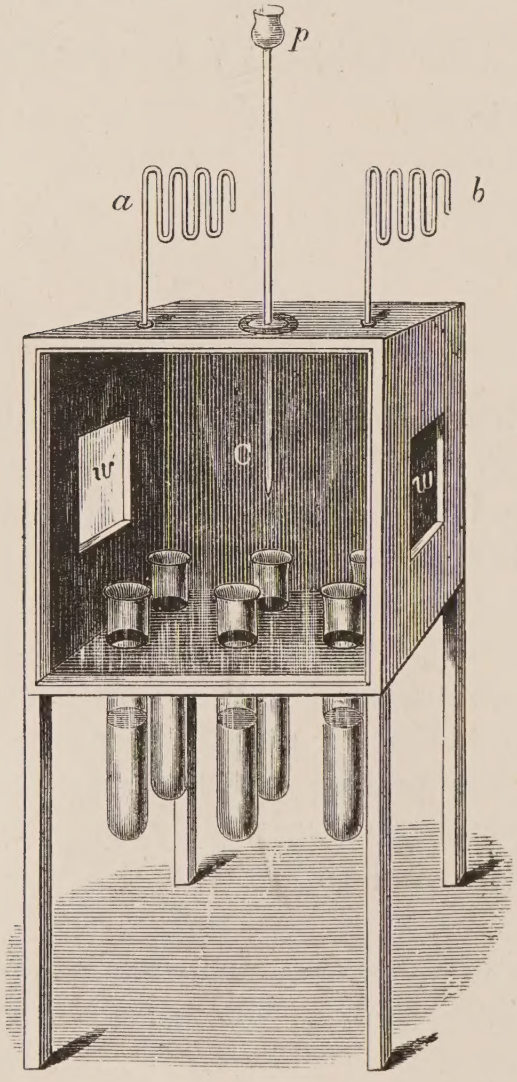
On the 13th of January, 1876, I had the honour of submitting to the Royal Society some account of an investigation in which the power of atmospheric air to produce life in organic infusions and its power to scatter light were shown to go hand in hand. The 'scattering' was proved to be due, not to the air itself, but to foreign matter suspended in the air. It was moreover proved that air placed under proper conditions went through a process of self-purification, and that, when this purification was visibly complete, the power to scatter light and to generate life disappeared together.

The form of the experiments here referred to was, it will be remembered, as follows:—Wooden chambers were constructed with glass fronts, side windows, and back doors. Through the bottoms of the chambers test-tubes passed air-tight, their open ends, for about one-fifth of the length of the tubes, being within the chambers. Provision was made for a connexion through sinuous channels between the outer and the inner air.

[1] Philosophical Transactions, Part I., 1877.

The chambers, being closely sealed, were permitted to remain undisturbed for a few days. The floating matter of the internal air gradually subsided, until at length an intensely luminous beam failed to show its

FIG. 11.



track within the chamber. Then, and not till then, were the infusions introduced, by means of a pipette passing through the top of the chamber. After their introduction, they were boiled in an oil or brine-bath [1]

[1] From the fact of their being boiled in oil or brine, Professor

for five minutes, and afterwards placed permanently in a warm room.

The annexed woodcut (fig. 11), taken from the 'Proceedings' of the Royal Institution, shows a chamber with its six test-tubes, its side windows *w w*, its pipette c, and its bent tubes *a b*, which connect the air of the chamber with the external air.

In upwards of fifty chambers thus constructed, many of them used more than once, it was, without exception, proved that a sterilized infusion in contact with air shown to be self-cleansed by the luminous beam remained sterile. Never, in a single unexplained instance, did such an infusion show any signs of life. That the observed sterility was not due to any lack of nutritive power in the infusion, was proved by opening the back door and permitting the uncleansed air to enter the chamber. The contact of the floating matter with the infusions was invariably followed by the development of life. Numerous examples of these results were placed before the Fellows of the Royal Society at their Meeting on the 13th of January, 1876.

Prior to the date here referred to, great public interest had been excited, and, I may add, considerable scientific uncertainty had been produced in reference to this subject, both in England and America, by the writings of Dr. Bastian. These writings consisted, in part, of theoretic considerations and reflections, not new, but sometimes very ably stated, based on the general doctrine of Evolution; and, in part, of very pungent criticisms of those who, though believers in Evolution, declined to accept the writer's programme

Cohn has inadvertently inferred that the infusions themselves were raised above their boiling-points. The tubes being open, the temperature of ebullition is of course independent of the source which provokes it.

of its operations.[1] Passing over both theory and criticism, I thought it wise to fix upon certain well-defined statements of fact which lay at the basis of the weighty superstructure raised by their author, and to bring these statements to the test of strict experiment.

Thus it was affirmed 'that boiled turnip or hay-infusions exposed to ordinary air, exposed to filtered air, to calcined air, or shut off altogether from contact with air, are more or less prone to swarm with *Bacteria* and *Vibriones* in the course of from two to six days.'[2] I resorted accordingly to filtered air, calcined air, and to infusions withdrawn from air, but failed to discover the alleged 'proneness' to run into living forms. It had also been affirmed that infusions of muscle, kidney, or liver, placed 'in a flask whose neck is drawn out and narrowed in the blowpipe flame, boiled, sealed during ebullition, and kept in a warm place, swarmed after a variable time with *Bacteria* and allied organisms.'[1] I resorted to such flasks, employing infusions of fish, flesh, fowl, and viscera, and on the 13th of January was able to place before the Royal Society one hundred and thirty flasks, every one of which negatived the foregoing statement.

Two objections were subsequently urged against these results. The infusions, it was contended, were not sufficiently concentrated, nor were the temperatures sufficiently high. Both these objections were met by the statement that forty-eight hours' exposure under the same circumstances to common air sufficed to fill these same infusions with life. Beyond this, however, I was able to show that the temperatures employed by me were exactly those which had previously been found

[1] See 'Evolution, or the Origin of Life,' pp. 168, 169.

[2] 'Evolution,' p. 94.

[3] Transactions of the Pathological Society, 1875, p. 272.

most effectual by the writer who urged the objection. Other temperatures, higher than any previously employed, were at the same time said to ensure spontaneous generation. I exposed my infusions to these newly-discovered efficient temperatures, but found that they remained as barren as before.

With regard, moreover, to the question of concentration, it was shown that, owing to their gradual vaporization, the infusions used by me were probably unequalled in strength by those employed by any previous investigator. Some of these infusions remain with me to the present hour. Concentrated by twelve months' slow evaporation, and reduced to one-fifth of their primitive volume, they still exhibit the purity of distilled water.

These results proved beyond a doubt that in the atmospheric conditions existing in the laboratory of the Royal Institution during the autumn, winter, and spring of 1875-6, five minutes' boiling sufficed to sterilize organic liquids of the most diverse kinds. Among these may be mentioned urine in its natural condition, infusions of mutton, beef, pork, hay, turnip, haddock, sole, salmon, cod-fish, turbot, mullet, herring, eel, oyster, whiting, liver, kidney, hare, rabbit, barn-door fowl, grouse, and pheasant. Once properly sterilized, and protected afterwards from the floating matter of the air, not one of these putrescible infusions ever manifested the power of generating by its own inherent energy putrefactive organisms of any kind.

§ 2. *Experiments of Pasteur, Roberts, and Cohn.*

During the investigation just referred to I confined myself for the most part to animal and vegetable juices in their natural condition—that is to say, extracted

by distilled water, and not rendered artificially acid, neutral, or alkaline. I had occasion, however, to repeat among others some of the very remarkable experiments on superneutralized hay-infusions described by Dr. William Roberts in his excellent paper in the Philosophical Transactions for 1874. These experiments I could not corroborate; for while in his hands such infusions sometimes required three hours' boiling to sterilize them, in mine they behaved like other infusions, and were sterilized in five minutes.

In the abstract of the investigation communicated to the Royal Society on the 13th of January, 1876, I mentioned this discrepancy, and pointed out its possible cause.[1] But the largeness of the question, which had been long previously raised by M. Pasteur, and the limitation of my time, led me to postpone it. This postponement is mentioned at the conclusion of my paper in the Philosophical Transactions for 1876, where the discrepancy referred to is not at all discussed.

In his celebrated paper, 'Sur les corpuscules organisés qui existent dans l'Atmosphère,' published fifteen years ago,[2] M. Pasteur first announced that while acid infusions had their germinal life destroyed by a temperature of 100° C., a temperature over 100° was needed to produce the same effect in alkaline infusions. In his 'Études sur la Bière,' published in the early part of 1876, he repeats and illustrates this statement. Vinegar he finds has the organisms which decompose it destroyed by a temperature of 50° C. Wine is rendered unchangeable by a slightly higher temperature. Beerwort without hops requires a temperature of 90° C. to sterilize it, and milk a temperature of 110°. Fresh urine has its organisms destroyed at a temperature of

[1] Roy. Soc. Proc. vol. xxiv. p. 178.

[2] Annales de Chimie, 1862, vol. lxiv.

100°, while a higher temperature is needed when the urine has been neutralized by carbonate of lime.[1] The resistance of alkalized urine to sterilization is therefore by no means a new announcement.[2]

On my return from Switzerland in 1876 the experiments on alkalized hay-infusions were resumed; and soon afterwards Professor Cohn, of Breslau, so highly distinguished by his researches on *Bacteria*, placed in my hands a memoir[3] which rendered it doubly incumbent on me to examine more strictly the grounds of my dissidence from Dr. Roberts. Professor Cohn is, on the whole, emphatic in his corroboration of Dr. Roberts,[4] having found, during a long and varied series of experiments with hay-infusions of divers kinds, that when the period of boiling did not exceed fifteen minutes organisms invariably appeared in the infusions after-

[1] 'Études sur la Bière,' p. 34.

[2] With regard to the different action of acid and alkaline liquids, I put the subject purposely aside with the view to its full investigation as soon as the first instalment of these researches had been published. I could find no adequate explanation of the alleged fact that germs are killed in an acid liquid, while they survive in an alkaline one of the same temperature; nor could the well-merited respect that I feel for M. Pasteur cause me to accept his explanation without further inquiry on my own account. In due time, therefore, I resolved to examine the question. Various experiments and explanatory views regarding it are recorded in the following pages.

[3] Beiträge zur Biologie der Pflanzen, July 1876.

[4] Professor Cohn gently censures me for taking exception to the cotton-wool plug, seeing that cotton-wool, even in my own experiments, has always proved a trustworthy filter. I did not, however, object to it as a filter, but on grounds which have in part, at all events, commended themselves to Professor Cohn himself. With reference to the method of Dr. Roberts he writes thus:—'The defect of this method consists in the difficulty of protecting the cotton-wool from accidental wetting by the infusion. The steam, moreover, which rises from the liquid penetrates the cotton-wool, and, through its partial condensation in the neck of the bulb, might readily charge itself with germs.'

wards. Sixty, eighty, and even one hundred and twenty minutes' boiling were found in some cases insufficient to sterilize the infusions. One marked difference, however, exists between Dr. Roberts and Professor Cohn. The former found five minutes' boiling sufficient to sterilize unneutralized hay-infusion, but one, two, and even three hours' boiling insufficient to sterilize superneutralized hay-infusion; while the latter noticed no difference of this kind, but found acid and neutral infusions equally resistant.[1]

§ 3. *Hay-infusions. Preliminary Experiments with Pipette-bulbs.*

I have now the honour to submit to the Royal Society an investigation which embraces among others the points here referred to, and which has proved far more difficult and laborious than I expected it would be.

On the 27th of September, 1876, a quantity of chopped hay was digested for three hours and a half in distilled water maintained at a temperature of 120° Fahr. The infusion was afterwards poured off, and its specific gravity reduced to the exact figure given by Dr. Roberts, viz. 1006. It was then filtered and slightly superneutralized. Precipitation occurred on the addition of the potash, and the infusion was boiled for five minutes to render the precipitation complete. It was then refiltered, and introduced into a series of bulbs of the same size and character as those described by Dr. Roberts, and called by him 'plugged bulbs.'[2]

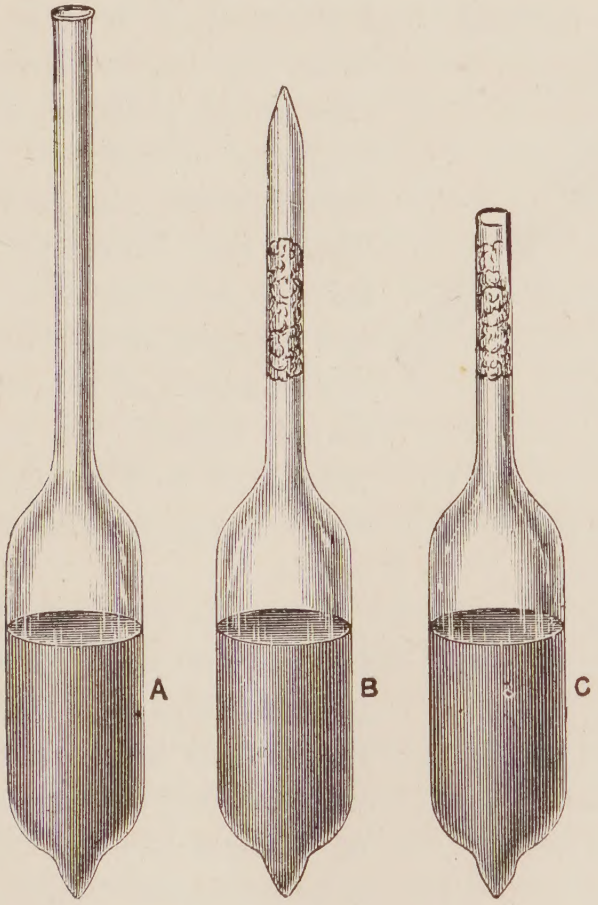
Each bulb was a cylinder about four inches high and upwards of an inch wide, with a long neck attached to

[1] 'Ein constanter Unterschied in der Zeitdauer zwischen sauren und neutralen Aufgüssen, wie ihn Roberts gefunden, trat in unseren Versuchen nicht hervor' (p. 259).

[2] Phil. Trans. vol. clxiv. p. 460.

it,[1] as shown at A, fig. 12. Two-thirds of the cylinder were occupied by the infusion. After the introduction of the latter, the neck of the bulb was plugged with cotton-wool, and hermetically sealed above the plug, as at B, fig. 12. The bulbs were afterwards plunged in water deep enough to cover their necks. The water was

FIG. 12.



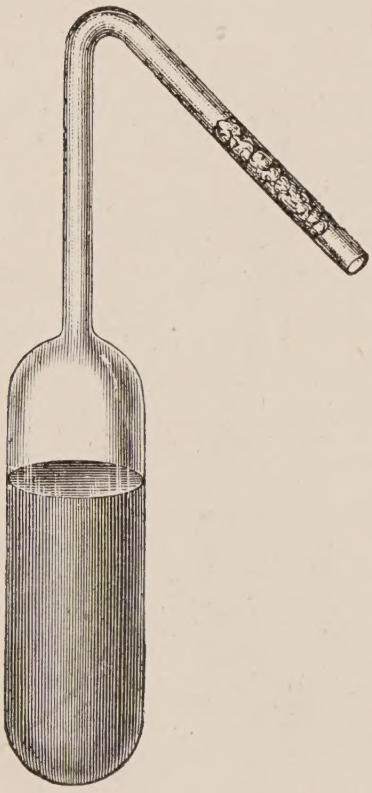
gradually raised to the boiling-point, and maintained at the boiling temperature for ten minutes. They

[1] I have called them 'pipette-bulbs' because they are formed by hermetically sealing one shank of a pipette, close to the bulb, leaving the other shank open for the introduction of the infusions. German pipettes, on account of their cheapness, were at first commonly used; but in cases of long-continued boiling, explosions were so frequent that bulbs of English glass of specially resistant quality were resorted to.

were then removed and permitted to cool; after which the sealed end of each neck was broken off by means of a file, its subsequent appearance being shown at c, fig. 12. The bulbs, protected by the cotton-wool plugs in the neck above them, were then exposed to a tolerably uniform temperature of about 90° Fahr.

At the same time two similar bulbs, charged with the same infusion, had their necks bent downwards, as in fig. 13, the inclined portion being plugged, so that no impurity could fall into the liquid from the cotton-wool. These two bulbs were boiled for five minutes in an oil-bath, and plugged while boiling with cotton-wool. They were then sealed behind the plugs and permitted to cool, their sealed ends being broken off afterwards.

FIG. 13.



On the 30th of September the infusion in all the straight-necked bulbs was turbid, while in the two bent-necked ones it was perfectly clear. On the 2nd of October the turbidity of the straight-necked bulbs had increased, while a fatty scum had formed on the surface of each. The two others were at the same time slightly but distinctly turbid.

My inference from this experiment was that in neither the straight-necked nor the bent-necked bulbs had the germs been wholly killed by the boiling. The difference between the results obtained with the respective bulbs arises from the different modes of mani-

pulation pursued in the two cases. For it is to be noted that a quantity of air, with its associated floating matter, was imprisoned above the infusion in every straight-necked bulb; that in the case of the two bent-necked bulbs this air had been in part displaced by steam, the air which entered on cooling being sifted by the cotton-wool plugs. To this difference of treatment is to be attributed the observed difference of deportment. Unlike the thick cloudiness of their neighbours, the turbidity of the bent-necked bulbs, though distinct, was barely sensible, and in none of them was any scum ever formed upon the surface of the infusion.

Examined microscopically, numerous Vibrios were found in the infusions of the straight-necked bulbs, many of them broken at the centre, with the two halves apparently trying to separate from each other. There were also numerous smaller *Bacteria*, very active and of various lengths. In the bent-necked bulbs a number of exceedingly small *Bacteria* were found, but no Vibrios.

The deportment of the hay-infusion employed in these experiments corroborates the results of Dr. Roberts and Professor Cohn.

On the 2nd of October another infusion of hay was prepared, and, after neutralization with caustic potash, was introduced into six pipette-bulbs with straight necks. The necks, being first plugged with cotton-wool, were afterwards sealed by the blowpipe. The infusions were maintained for ten minutes at the temperature of boiling water. Their sealed ends were afterwards broken off, and they were subjected, like the former ones, to a temperature of 90° Fahr.

Six other bulbs were charged at the same time with the same infusion; but, instead of being hermetically sealed, they were placed in an oil-bath, and boiled there

for five minutes. Before the ebullition ceased, the neck of each was stopped with a plug of cotton-wool.

Up to October 6th all the bulbs continued clear. On the 6th one bulb of the series last described became turbid, lighter in colour than its neighbours, and covered with a fatty scum. On the 7th one tube of the first series (boiled after the fashion of Roberts for ten minutes) also became turbid and exhibited the same fatty scum. The remaining ten bulbs maintained permanently their deep brown-sherry colour, their high transparency, and their perfect freedom from Bacterial life. They are still clear, though seven months have elapsed since their preparation.

In the great majority of these experiments the deportment of alkalized hay-infusion contradicts that observed by Dr. Roberts and Professor Cohn.

Six other pipette-bulbs, with their necks so bent and plugged with cotton-wool and asbestos that no impurity falling from the plug could reach the infusion, were also charged on the 2nd of October. Three of the bulbs, with their necks hermetically sealed, were maintained for ten minutes at the temperature of boiling water, the sealed ends being afterwards broken off. The three other bulbs were boiled in an oil-bath, and had their necks plugged before ebullition ceased. All six bulbs have remained perfectly transparent up to the present time.

Here, again, we have discordance between my results and those of Dr. Roberts and Professor Cohn.

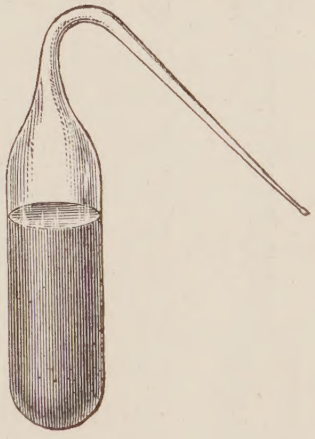
But on the 6th of October another infusion was prepared and neutralized, exactly in the same fashion as before. Five pipette-bulbs were charged with it; they were hermetically sealed and maintained at the boiling temperature for ten minutes. The sealed ends were afterwards broken off, and the bulbs exposed to a

temperature of 90° Fahr. On the morning of the 8th of October (that is to say, two days after their preparation) the infusion in every one of the bulbs was turbid and covered with scum.

Here once more we have perfect harmony between my results and those of Dr. Roberts and Professor Cohn.

On the 2nd of October, moreover, fourteen of our ordinary small retort-flasks with bent necks (shown in fig. 14) were charged with the neutralized hay-infusion. They were boiled for three minutes, and hermetically sealed whilst boiling. Some days afterwards one tube of the entire number was observed to have become lighter in colour and sensibly cloudy; but thirteen out of the fourteen remained unchanged in colour, brightly transparent, and entirely free from life.

FIG. 14.



Here the dissidence between my results and those of Professor Cohn, who also experimented with hermetically-sealed flasks, reappears.

Numerous other experiments with pipette-bulbs and retort-flasks were made at the time here referred to, but it is unnecessary to record them. Suffice it to say that, like those just described, some of them corroborated and some of them contradicted the results of Dr. Roberts and Professor Cohn.

§ 4. *Hay-infusions. Experiments with Cohn's Tubes.*

For reasons given by himself,[1] Professor Cohn deviated from the method of experiment pursued by Dr. Roberts, employing, instead of the pipette-bulbs, flasks, the nature of which will be understood from the following description. Let a zone of a common test-tube, about one-third of its length from its open end, be softened by heat, and let the softened glass be drawn out so as to form a tube of much narrower bore than the original test-tube. Thus modified, the tube would consist of an elongated bulb below and an open funnel above, both being connected by a narrow neck (see fig. 15). Professor Cohn filled the elongated bulb to about two-thirds of its volume with hay-infusion, plunged his bulbs in water, raised the water to ebullition, and continued the boiling for the required time. The tubes were then removed from their bath, and after being held open for a minute or two so as to allow the water condensed in their necks to evaporate, the funnel was plugged with cotton-wool.

FIG. 15.

Professor Cohn considers that all possibility of external contamination is here shut out.[2] By his method, therefore, I wished to check the results above

[1] Beiträge, July 1876, p. 256.

[2] 'Ehe ich über die Organismen berichte, welche sich in den gekochten Aufgüssen entwickelten, will ich bemerken, dass an eine nachträgliche Infection derselben durch von aussen nach dem Kochen eingeschleppte Keime bei unseren Versuchen nicht zu denken ist' (p. 259). I may remark that, with an atmosphere like that in which my recent experiments were conducted, there would be no chance of escape for an infusion thus handled.

described. Accordingly, on the 24th of October, I had four groups of Cohn's tubes (twelve in a group) carefully charged with two fresh infusions of two different kinds of hay. Each infusion was divided into two equal parts, one of which was neutralized and the other left in its natural acid condition. Twelve of the tubes were charged with one of the infusions neutralized, and twelve with the same infusion unneutralized. We will label this infusion A. Twelve other tubes were charged with the second infusion neutralized, and twelve with it unneutralized. We will call this infusion B. The forty-eight tubes were subsequently boiled for ten minutes in tin vessels containing water deep enough nearly to submerge them. Having proved by previous experiments that it was dangerous if not fatal to exactness to expose the infusions for one or two minutes to the air after their removal from the water, I took the precaution of plugging them first and removing them afterwards.

On the 28th of October (that is to say, four days after their preparation) several of the tubes containing the unneutralized infusion A were faintly but distinctly turbid and thinly covered with scum. The twelve neutralized tubes of the same infusion were at the same time perfectly clear. This retarding influence of the alkali has been of frequent occurrence in this inquiry. That it was simply a case of retardation was proved by the fact that, on the 30th of October, the twenty-four tubes, both neutral and acid, of infusion A were turbid and covered with scum.

On the same date the twelve neutralized tubes of infusion B were perfectly clear and without a trace of scum. Of the twelve unneutralized tubes three had given way, and a fourth yielded on the 31st. Four days later three of the neutralized tubes also yielded. The permanent state of matters was that eight out of

the twenty-four tubes charged with infusion B had become turbid, while sixteen of them remained perfectly clear. I do not doubt that the tardy infection of some of the tubes just referred to arose from external contamination, which is almost inseparable from the method of experiment.

Here, while infusion A *corroborated Professor Cohn, infusion* B *in substance contradicted him.*

§ 5. *Hay-infusions (in Closed Chambers).*

In dealing with hay-infusions I also fell back on the method of experiment which was found so effectual in 1875,[1] employing closed chambers in which the air had been permitted to cleanse itself by the gradual subsidence of its floating matter.

On the 3rd of October, 1876, my experiments with such chambers recommenced. Two of them, containing each three large test-tubes, were then charged with an infusion of hay accurately prepared according to the prescription of Dr. Roberts. Its specific gravity was 1006; it was superneutralized to the proper extent with caustic potash, but the period of boiling, instead of being three hours, was five minutes.

Examined from time to time for more than four months subsequently, the infusion in both chambers continued perfectly unchanged. It was free from suspended matter, free also from every trace of scum, maintaining for the light which passed through it a singular transparency.

Here, to a certainty, a period of boiling not amounting to one-twentieth of that required by Dr. Roberts, sufficed to destroy totally the power of generating life in an alkalized hay-infusion.

[1] Briefly described in the Introduction.

This result is in perfect harmony with *all* the results of last year. Chamber after chamber was then charged with infusions of hay, which were afterwards subjected to the boiling temperature for five minutes. In every chamber the infusion remained perfectly clear until purposely infected from without. There was no instance observed last year in which five minutes' boiling failed to sterilize hay-infusion, whether neutralized or unneutralized.

Thus, on the 26th of November, 1875, a group of three test-tubes was charged with hay-infusion of the same specific gravity and of the same degree of alkalinity as that found most resistant by Dr. Roberts. They were protected by glass shades, the air within the shade being calcined by an incandescent platinum wire in the manner described in the last essay.[1] The tubes were boiled for five minutes, the subsequent intrusion of contaminated air being prevented by a ring of cotton-wool. Thirteen months afterwards the infusion, greatly concentrated by evaporation, exhibited its pristine deep transparency. A second similar group of tubes was charged with alkalized hay-infusion on the 27th of last January, and on the 5th of December (that is to say, after a period of more than ten months) the infusion was found perfectly clear.

A number of hermetically-sealed tubes charged with the same infusion, and boiled for only three minutes, have maintained for more than a year both their primitive transparency and their water-hammer sound. Thus many of the earliest experiments of the present year, and the whole body of last year's experiments, are in complete harmony with each other.

This harmony was, however, disturbed by some of the foregoing experiments with bulbs and tubes, and it

[1] Phil. Trans., vol. clxvi. p. 50.

was soon to be further disturbed by experiments with closed chambers. On the 6th of October, 1876, for example, an infusion was got ready in strict imitation of that prepared on the 3rd; it was of the same specific gravity, it was alkaline to the same degree, and it was introduced in the same manner into a chamber of three tubes; but whereas the infusion of the 3rd remained intact for months, and would have remained so indefinitely, a week had not elapsed before every tube of this new infusion was turbid and covered with fatty scum.

§ 6. *Desiccation of Germs. New Hay and old.*

In his work entitled 'Evolution, and the Origin of Life,' Dr. Bastian affirms, with repeated emphasis, that living matter is unable to maintain its life when exposed to a temperature even below that of boiling water. He refers to the scalding of the hand and other destructive effects, and also to the action of boiling water on eggs. He also refers to the experiments of Spallanzani on seeds, and extends the results observed with living matter of these special kinds, to living matter generally. 'It has been shown,' he writes,[1] 'and is believed by the great majority of biologists, that the briefest exposure to the influence of boiling water (212° F.) is destructive of all living matter.'

More than ten years ago an extremely significant observation directly bearing upon this subject was made by the wool-staplers of Elbœuf, in France. They were accustomed to receive dirty fleeces from Brazil, and among other matters entangled in the wool were the seeds of a certain plant called *Medicago*. It had been repeatedly found by the wool-cleaners that these seeds sometimes germinated after a period of four hours'

[1] 'Evolution,' p. 46.

boiling. The late M. Pouchet repeated the experiment. He collected the seeds, boiled them for four hours, and sowed them afterwards in proper earth. To his astonishment they proved fruitful. He then closely examined the boiled seeds, and found the great majority of them swollen and disorganized; but amongst these ruined seeds he observed others which had refused to imbibe the water or to swell or break up in any way. These he carefully picked out, and sowed them and their neighbours separately in the same kind of earth. The swollen seeds were incapable of germination, while the unaltered ones rapidly gave birth to a crop. This was the only instance of such resistance known to Pouchet when he communicated the fact to the Paris Academy of Sciences.

The observation here described stands recorded in the 'Comptes Rendus' for 1866, vol. lxiii. p. 939. It is not difficult, indeed, to see that the surface of a seed or germ may be so affected by desiccation and other causes as practically to prevent contact between it and a surrounding liquid.[1] The body of a germ, moreover, may be so indurated by time and dryness as to resist powerfully the insinuation of water between its constituent molecules. It would be difficult to cause such a germ to imbibe the moisture necessary to produce the swelling and softening which precede its destruction in a liquid of high temperature.

In my last paper I made some remarks upon this subject;[2] and in relation to our present experiments,

[1] In this connexion a remark of Dr. Roberts regarding the resistance of chopped green vegetables merits quotation. 'The singular resistance of green vegetables to sterilization appears to be due to some peculiarity of the surface, perhaps their smooth glistening epidermis, which prevented complete wetting of their surfaces.'

[2] Phil. Trans., vol. clxvi. p. 60.

the influence of drying and hardening was brought home to me by the fact that in all the foregoing cases the infusions which five minutes' boiling proved sufficient to sterilize *were, without exception, derived from fresh hay mown in* 1876, *while the infusions which five minutes' boiling failed to sterilize were derived, without exception, from old hay mown either in* 1875 *or some previous year.*

In the earlier experiments of the present inquiry this distinction between old and new hay came most clearly and definitely out. The result was subsequently blurred by circumstances which it required time and labour to unravel, and which will require patience on the reader's part if he would thoroughly follow them. They will, however, throw far more light upon the real character of these inquiries, and do more to reconcile the discords to which researches on spontaneous generation have given birth, than if every experiment had been a success unshaded by doubt.

§ 7. *Hay-infusions. Further experiments with Closed Chambers.*

With a view to probing to the uttermost this question of drying and hardening, on the 6th of October an extensive series of experiments with closed chambers was begun. Three different kinds of hay were employed:—1st, Old hay, sent to me by Lord Claud Hamilton, from Heathfield, Sussex;[1] 2nd, new hay from Heathfield (both, it may be stated, from a somewhat ungenerous soil); 3rd, new hay purchased in London, and artificially dried for some days upon a sand-

[1] After the possible influence of hard drying and hardening had suggested itself, I purposely introduced old hay from various localities into the laboratory.

bath. For these experiments eleven closed chambers were prepared, as I wished every result to be based as far as possible upon the testimony of two chambers. On the 6th of last October they were carefully charged with the infusions, the period of boiling afterwards being five minutes.

Two chambers were devoted to the acid and two to the alkalized infusion of old hay. Two chambers were also devoted to the acid and two to the alkalized infusion of dried hay. Two chambers were finally devoted to the alkalized and one to the natural acid infusion of new Heathfield hay.

Examined from day to day, differences were soon observed, not only between the different infusions, but also between different chambers containing the same infusion. Thus every tube of both the chambers containing the neutralized infusion of old hay became turbid, but the three tubes of the one chamber were loaded in four days with a fatty scum, while the tubes of the other chamber remained for ten days perfectly free from scum. The two chambers containing the acid infusion of old hay exhibited similar differences. Every tube in both of them became turbid; but in one of them the infusion was scumless throughout, while in the other each of the three tubes was heavily laden with scum.

The two chambers containing the alkalized infusion of dried London hay had all their tubes turbid and covered with scum. In the case of the acid infusion of dried hay, the tubes of one of the chambers became turbid, while the tubes of the other chamber remained clear.

The two chambers of alkalized new Heathfield hay-infusion were also in disaccord. In the one chamber all three tubes became turbid and covered with scum,

while in the other chamber the three tubes remained sensibly clear and free from scum. Nor did the three tubes of the single chamber charged with the new Heathfield acid infusion present the same appearance; for while one tube became thickly turbid, the other two remained perfectly pellucid.

Amid this confusion, the only point worth dwelling on is, that while no single case of escape occurred with the old-hay infusion, whether acid or neutral, with the infusions of both dried and undried new hay a certain percentage of the tubes remained sterile.

Reflection on these results naturally drew suspicion upon the chambers. They had been used before, and, though carefully cleansed, some unobserved source of infection may have clung to them. This, at all events, seemed the most rational way of accounting for the differences observed between samples of the self-same infusion placed in different chambers. Hence my desire to expose a fresh series of infusions in chambers which had never been used before.

Six new ones were therefore constructed, each of them containing six tubes. These were charged on the 3rd of November with infusions of old London hay, old Heathfield hay, new London hay, and dried London hay. Two chambers were devoted to each infusion, which in the one chamber was neutralized and in the other unneutralized.

The six tubes in each chamber were arranged in two rows of three tubes each. Those nearest to the glass front were called the front tubes, the others the back tubes. The infusion intended for the unneutralized chamber was unboiled before its introduction into the three back tubes, and boiled in those tubes for five minutes afterwards; the infusion for the front tubes was

boiled for fifteen minutes before introduction and for five minutes afterwards. These differences in the mode and period of boiling were adopted to ascertain whether they had any influence on the subsequent development of life. In the case of the neutralized chambers, the infusion for the three back tubes was boiled for fifteen minutes outside before neutralization, and five minutes in the chamber after neutralization. The infusion for the three front tubes was boiled fifteen minutes outside after neutralization, and five minutes afterwards in the chamber. If the potash used for neutralization carried germs into the infusion, the difference between five and twenty minutes in the period of boiling might, it was thought, declare itself in the subsequent phenomena.

Four days after its introduction the old Heathfield acid infusion was found turbid throughout and covered with scum. The scum and turbidity were sensibly the same in all the tubes, though the period of boiling varied from five to twenty minutes. On the same day the neutralized infusion of the same hay was perfectly brilliant and free from scum. Three days subsequently, however (that is to say, on the 10th of November), the neutralized tubes also became turbid and covered with scum.

The salient fact here to be noted is, that in neither the neutral nor the acid chamber did a single tube of the old Heathfield hay-infusion maintain its primitive clearness and freedom from scum.

The old London hay behaved substantially as the old Heathfield hay, no single tube escaping either in the neutralized or the unneutralized chamber.

The dried new London hay comes next. A week after its introduction every one of the six tubes containing the acid infusion was turbid and coated with scum. In the neutralized chamber, on the contrary,

two only of the back tubes gave way, the third back tube and the three front tubes remaining clear.

On the 3rd of November, moreover, a new chamber of six tubes was charged with an infusion of new London hay. Three of the tubes were neutralized and three unneutralized. Both infusions were introduced into the chamber unboiled, and were boiled afterwards for five minutes. In a week all the tubes had given way, becoming turbid in the same degree and covered to the same extent with scum. The newness of the hay had failed to secure the sterility of the infusions.

Nothing of this kind occurred in the experiments of last year. It was then found that hay-infusions of all kinds were uniformly sterilized by five minutes' boiling.

Guided by such hints as the experiments furnished, I continued to work. On the 4th of November four closed chambers of three tubes each were charged with infusions of old and new Heathfield hay—two chambers with the one, and two chambers with the other. One chamber of each pair contained a neutralized, the other an unneutralized infusion, and the time of boiling was ten minutes. Six days subsequently the infusion of new hay, both neutralized and unneutralized, was found perfectly unchanged. Of the old-hay infusion, on the other hand, only one of the six tubes escaped. The three acid tubes became completely turbid, while two out of the three neutral ones fell into the same condition.

§ 8. *Experiments with Soaked Hay.*

Pondering still further on the influence of drying and hardening, and recognizing the necessity of not only wetting but also softening the germs, the thought

occurred to me of soaking the hay for some days prior to digesting it. Old London hay was accordingly chopped up and placed in three glass vessels—one containing distilled water, another acidulated water, and a third alkalized water. The superior extractive power of the alkalized liquid was at once manifest; it rapidly assumed a dark colour. The distilled water came next, yielding a colour less deep than that of the alkalized, but more deep than that of the acidulated water. The alkaline and distilled-water infusions emitted a rich odour of hay, while the smell of the acid infusion was very faint, and not like that of hay. The hay was permitted to soak from the 8th to the 11th of November. It was then digested for three hours in the same liquid at a temperature of 120° F., boiled, filtered, and introduced into the closed chambers, where it was reboiled in each case for five minutes.

Prior to digesting the hay in the liquid in which it had been soaked, *Bacteria* had developed in swarms. These, of course, were killed by the boiling, and they were not entirely removed by the filtration. The alkaline infusion, indeed, though filtered repeatedly, was sufficiently turbid to prevent the flame of a candle placed behind the tubes containing it from being seen. The same to a less extent was true of the distilled-water infusion. This latter had been divided into two portions, one of which was accurately neutralized, and the other left unneutralized, a separate chamber being devoted to each.

From the 11th to the 18th of November the only change observed in any of the infusions was in the direction of increased transparency. They all became clearer with time, the distilled-water infusions becoming particularly clear and brilliant at the top. After two or three days' quiet the alkaline infusion allowed a flame

placed behind it to be seen of a deep red. The acidulated-water infusion remained entirely unchanged; but this is not worth dwelling on, for in this case, even when exposed to the common air, the infusion resisted infection for a considerable time.

In no case was the fatty scum which had been already so frequently observed formed in any one of the tubes. Some change inimical to the particular organisms which produce this scum must have been caused by the soaking of the hay.

Examined microscopically on the 18th of November these infusions, I thought, exhibited undoubted evidences of Bacterial life. Bacterial forms were unquestionably there in considerable numbers, more particularly in the sediment at the bottoms of the tubes. Nor do I now see any valid grounds for doubting the presence of life; but I was warned against drawing too hastily the conclusion which first prompted itself, by boiling an infusion swarming with active *Bacteria*, and submitting the liquid after cooling to microscopic examination. Here also the dead Bacterial forms were preserved, and it was extremely difficult to distinguish their motions, which were certainly Brownian motions, from those observed in the protected infusions of soaked hay.

The experiment was thought worth repeating. On the 16th of November accordingly chopped bundles of old Heathfield hay and new Heathfield hay, and of old London hay and new London hay, were placed in glass dishes containing distilled water, and were thus soaked until the 18th. They were then moved from the lower laboratory, and taken, with their glass covers, to a distant room at the top of the Royal Institution. Here the four specimens of hay were digested for three hours at a temperature of 120° Fahr. They were filtered, boiled, refiltered, some of them through 100 layers of

filter-paper; after which they were introduced into four closed chambers of six tubes each, and then boiled for five minutes.

On the 20th of November the infusions in all the chambers appeared to be as free from organisms as at first. The new Heathfield and the new London hay-infusions in their respective chambers had their somewhat turbid columns surmounted by an exceedingly clear zone of liquid, due, I should consider, to the mechanical subsidence of the particles, had not subsequent experience taught me to regard this appearance as a sign of life.

On the 23rd scum had begun to gather on every tube of the case containing the infusion of old Heathfield hay. On the 30th this scum continued, but there was no trace of it in any of the chambers containing new Heathfield hay, new London hay, and old London hay. These infusions were all somewhat turbid; but the turbidity differed very little from that exhibited when the infusions were prepared.

I spent a good deal of time over these infusions of soaked hay, both with the microscope and otherwise, but the recorded observations would not add materially to our knowledge. I therefore dismiss them with the remark that their general drift was in favour of the idea that the extraordinary resistance to sterilization manifested by the old-hay infusions is the result of hardening and desiccation. The foregoing observations, however, have been noted, more with the view of indicating my line of thought than of claiming for them any value whatever as a demonstration.

§ 9. *Infusions of Fungi.*

Turning from hay to substances in which germs, if they existed, could not be desiccated, I felt pretty sure that infusions of such substances would be unable to resist the boiling temperature. To test the correctness of this view the following experiments were made :— Three different kinds of fungi (red, black, and yellow) were gathered in Heathfield Park on the 13th of October, and digested separately in London on the following day. Three tubes of a closed chamber containing six tubes were charged with the red-fungus infusion and three with the black, while a second chamber of three tubes was charged with the yellow-fungus infusion. They were all boiled for five minutes after their introduction into the chambers.

For two or three days all the infusions continued clear; but they subsequently broke down, every tube of the nine becoming turbid with organisms and covered with scum.

Examined microscopically on the 8th of November the red-fungus infusion was found charged with a multitude of spore-like bodies, massed in some places continuously together, in others floating freely in the liquid. Among these ran long filaments, dotted with spore-like specks from beginning to end. There was a considerable number of Vibrios in one of the tubes. The black-fungus infusion contained a mixed population of Vibrios and *Bacteria* with spore-filled filaments. Swarms of *Bacteria* were observed in the red-fungus infusion.

Suspicious of the chambers in which these infusions had been exposed, I had three new ones constructed and provided with new tubes. A fresh supply of fungi was sent to me from Heathfield, a tree fungus being,

however, substituted for the black one used in the former experiments. On the 1st of November the three infusions were very carefully introduced into three chambers, a chamber being devoted to each infusion. I thought it advisable to vary the period of subsequent boiling. One tube of the yellow fungus was therefore boiled for five, one for ten, and one for fifteen minutes; but as it was difficult to save the infusion from waste when the boiling was long continued, one tube of each of the other two infusions was boiled for five minutes, and the other two for ten. Tubes charged with the respective infusions were exposed at the same time to the common air.

In two days the outside tubes containing the red- and yellow-fungus infusion became turbid and covered with the fatty scum so prevalent in our laboratory this year. No scum had formed on the surface of the exposed tree-fungus infusion, which, to casual observation, appeared quite black. Closer scrutiny, however, showed that it transmitted the deepest red of the spectrum, and was apparently quite free from floating matter. It changed rapidly during the night of the 3rd, and on the morning of the 4th of November the bottom of this tube was found laden with a heavy dark-brown precipitate, while numerous dark-brown flocculi floated in the liquid overhead, which had become almost as clear and colourless as water. Under the microscope the dark-brown mass resolved itself into confused moss-like patches and long cylindrical sheaths dotted throughout with small dark specks. These filaments with spore-like specks have been of very frequent occurrence in this inquiry.

The deportment of the closed chambers was as follows:—1. Yellow fungus: the liquid in the three tubes remained perfectly and permanently clear and without

a trace of the scum which loaded the infusion outside. 2. Red fungus: one of the three tubes became thickly turbid, while the two others maintained their pristine brilliancy. 3. Tree fungus: one of the tubes became thickly turbid, the two others remained permanently clear.

I asked myself why should one tube of the red-fungus give way and the others remain intact? The answer seemed at hand. The turbid tube had been boiled for only five minutes, while the clear ones had been boiled for ten. On consulting the adjacent chamber this possible explanation was blown to the winds, for here the turbid tube had been boiled for ten minutes, while its untainted neighbour had been boiled for only five.

Thus, although the more careful repetition of the experiments did not secure every tube from infection, the escape of seven out of nine tubes entirely destroys the presumption of spontaneous life-development which the first experiments might suggest to some minds.

Wishing to observe more attentively the action of common uncleansed air upon boiled fungus-infusions, a tray of 100 tubes was charged with them on the 14th of October. Thirty-five tubes were filled with black, thirty-five with yellow, and thirty with red-fungus infusion. On the 16th of October every one of the yellow-fungus tubes was turbid and covered with a thick, coherent, cobweb-like scum. The surfaces of the black-fungus tubes were also sprinkled with spots of white scum. Turbidity was the only change observed in the red-fungus tubes. They were wholly free from scum.

Examined microscopically on the 2nd of November the yellow-fungus tubes were for the most part found swarming with exceedingly small and active *Bacteria*; the red-fungus tubes also swarmed with *Bacteria*, some beaded Vibrios being mingled with them. In many of the tubes examined galloping monads appeared, attain-

ing an astounding development in the black-fungus infusion. Patches of moss-like matter would appear here and there in the field of the microscope; and it was no uncommon thing to see from ten to twenty monads nestling and quivering in this 'moss,' and darting actively in and out of it. They put me in mind of frogs amid their spawn; and as I looked at them my belief in the animality of the one was almost as strong as in that of the other. Almost every patch of spawn-like matter had its colony. In some cases hardly any thing but monads was to be seen; but in others the crowding of active Vibrios was so great that the monads wholly retreated from the field.

§ 10. *Infusions of Cucumber, Beetroot, &c.*

The fungi having disappeared on the approach of winter, I turned to cucumber and beetroot, not expecting that their sterilization would offer any difficulty. Two closed chambers were accordingly prepared, left for the proper time in quietness, and on the 7th of November were charged, the one with the cucumber- and the other with the beet-root infusion. In a few days the infusions in both chambers broke down, first losing their transparency and afterwards loading themselves with fatty scum. Thus perplexities accumulated.

On the 18th of November twenty-four Cohn's tubes[1] were charged with infusions of cucumber, beetroot, parsnep, and turnip, six tubes being devoted to each infusion. They were placed in a vessel of cold water, raised gradually to the boiling-point, and maintained at the boiling temperature for ten minutes. Before their removal from the hot liquid they were one and all plugged with cotton-wool.

[1] See § 4.

On the 30th of November all the infusions were thickly turbid throughout and heavily coated with scum.

From some of the precautions already mentioned it may be inferred that before this point of the inquiry had been reached, I had begun to suspect the atmosphere in which I worked. Hay of various kinds, both old and new, had been exposed and shaken about in the laboratory, the air of which doubtless contained multitudes of spores which diffused and insinuated themselves everywhere. So, at all events, I reasoned. On the 20th of November, therefore, I had infusions of cucumber, beetroot, parsnep, and turnip prepared, far from the laboratory, in one of the highest rooms of the Royal Institution, and introduced into four new chambers of three tubes each. I deemed the precaution of preparing the infusions and introducing them in the distant room sufficient. Accordingly, when the chambers were charged they were carried down, and the infusions boiled in the laboratory.

Two days afterwards the parsnep alone remained clear. This, however, was only a respite, for a day or two subsequently it fell into the condition of its neighbours. On the 30th of November both turnip- and parsnep-infusions were turbid throughout, and laden at the surface with thick fatty scum. The cucumber was also heavily laden with scum, which sent long streamer-like filaments into the subjacent liquid. The beetroot agreed with the others in becoming turbid, but differed from them in remaining free from scum. In no case last year did turnip-infusion show the deportment here described. Knowing, then, from multiplied experiments, that turnip possessed no inherent power of life-development, the conclusion was irresistible that its present behaviour, and with it the behaviour of cucumber, beetroot, and parsnep, were due to infection from without.

I once more tried removal to a distant room, with the added precaution of not only introducing the infusions into the chambers upstairs, but of boiling them there. It had been noticed that when the test-tubes were withdrawn from the oil-bath, and the discharge of steam into the chambers ceased, a somewhat violent entrance of the air into the cooling chamber was the consequence. To sift such air of its germs, both the funnel of the pipette and the open ends of the bent tubes were carefully stopped with cotton-wool. The wool was never removed from the funnel, and it was not removed from the bent tubes until the chamber had thoroughly cooled. The same vegetables were operated on, viz. cucumber, beetroot, turnip, and parsnep. On the 25th of November four chambers were charged with the infusions. On the 30th they were one and all covered with a layer of deeply pitted and corrugated fatty scum. Thus far, then, I was defeated in my efforts to escape contamination.

During these experiments a fact was observed which repeated itself afterwards in other instances. Samples of the different infusions were always exposed to the common air beside their respective chambers, and in general these outside samples became turbid and covered with scum a day or so before the interior tubes gave notice of breaking down; but here, in the case of the turnip, the outside tube continued pellucid and free from life for some time after the inside ones had become turbid with organisms. How could this be? The case of my two trays placed one above the other last year [1] suggested itself to my memory. In regard to life-development it was then found that the lower tray was always in advance of the upper one. As pointed out

[1] Phil. Trans., vol. clxvi. p. 68.

at the time, the absence of agitation which permitted the germs to sink into its tubes was the cause of the quicker contamination of the lower tray. No other cause appeared to me assignable in the present instance. By some means or other germs had insinuated themselves into my closed chamber, where the tranquillity of the air permitted them to sink into the infusion, and thus produce effects in advance of those produced by the unquiet air outside. So, at all events, I reasoned.

But how could the germs get into the chamber? I could, at the moment, fix only upon one way. The weather had changed from warm to cold and from cold to warm. This genial outside temperature sometimes caused the air surrounding the infusions to rise to upwards of 90° Fahr., and we had often to work in this heat. To moderate it, I sometimes partially turned off the gas, thus lowering the temperature of the room 10° or more. The contraction of the air within the closed chambers followed as a matter of course, and the bent tubes being open, I thought the entrance of the external air might be sufficiently rapid to carry germs along with it.

A new chamber of six tubes was therefore prepared upstairs, three of its tubes being charged with cucumber- and three with turnip-infusion on the 27th of November. The pipette funnel and the bent tubes were plugged above with cotton-wool, which was not removed from them afterwards. I took care, moreover, not to alter the gas-stoves in any way. My care was nugatory. In three days every tube of the six was laden with life. Another chamber of six tubes, charged on the 30th of November with cucumber-infusion, and two additional ones prepared on December 1st, shared the same fate.

Slices of cucumber were next digested for three

hours; the infusion was filtered, boiled, and such precipitated matter as appeared on boiling was removed by refiltering. The liquid thus prepared was introduced into five thick glass tubes, which were hermetically sealed, placed in a cold oil-bath, gradually heated to 230°, and maintained at that temperature for a quarter of an hour. The tubes being removed and permitted to cool, the infusion was introduced into a chamber of six tubes, and boiled there for five minutes.

The previous superheating of the infusion did not even retard the development of life, for in less than two days every tube in the chamber swarmed with *Bacteria*. Thus far, then, every attempt at a solution was unsuccessful.

But why, it may be asked, attempt such solutions? Was it not mere prejudice against the doctrine of spontaneous generation that prevented me from frankly submitting to the apparent logic of facts, and admitting the experiments just recorded to be a demonstration of the doctrine? By no means. The only prejudice I feel is the wholesome repugnance to accepting momentous conclusions on insufficient grounds. Hume's celebrated argument has its application here. Taking antecedent experience fully into account, it was far easier for me to believe my knowledge imperfect, or my present work erroneous, than to believe the doctrine of spontaneous generation true.

§ 11. *New Experiments on Animal Infusions. Contradictory results.*

In the course of this inquiry I was continually reminded of my experiments in 1875, when the most complete immunity from Bacterial or fungoid life was so readily secured. I had operated many times with

turnip, never finding the least difficulty as to its sterilization. It is certain that the care bestowed in preparing the turnip-infusion on the 20th of November, 1876, was greater than that bestowed upon the same infusion in 1875. But whereas the latter was invariably sterilized by five minutes' boiling, remaining afterwards as pellucid as distilled water, the former, three days after its preparation, became thickly turbid and swarming with life. I extended the present inquiry to other substances whose deportment was familiar to me last year, some of whose infusions, indeed, still remain with me as clear as they were on the day of their preparation.

On the 1st of December, for example, infusions of beef, mutton, pork, herring, haddock, and sole were prepared, and introduced into six closed chambers, each containing three tubes. On the 5th of December the pork, beef, mutton, and haddock were all covered with a fatty corrugated scum. A second chamber, containing artichoke-infusion, prepared at the same time, was found on the 5th more turbid than any of the animal infusions, and equally covered with scum. In the animal infusions, indeed, the body of the liquid underneath the scum maintained a surprising brilliancy, the development of life being confined to the layer in immediate contact with the atmospheric oxygen.

On the 5th of December the herring- and sole-infusions were both clear; but this was only a respite, for on the 6th white spots appeared on the latter, which extended until they covered the whole surface. The herring-infusion remained clear for a week, after which small specks began to appear on its surface. They never reached the development of the scum which coated the other infusions. It sometimes occurred to me that the oil of this fish exercises a certain antiseptic action.

Last year I preserved infusion of herring perfectly pellucid for months, even in a chamber so leaky that the light could be seen through its chinks. I had, moreover, no failure with any of the animal infusions here enumerated. Last year they all remained sweet and clear; this year, with far greater precaution, I failed to protect any of them from putrefaction. Reflection on these results, renders, I think, but one conclusion possible to the scientific mind. It will be loth to assume that mutton, beef, pork, haddock, herring, and sole had totally changed their natures, and contracted qualities and powers in 1876 which they did not possess in 1875. But if the origination of the observed life be denied to the infusions themselves, there is but one other source to which it could be referred, namely, atmospheric contamination.

It became, indeed, more and more obvious to me that, in consequence of increased vitality or virulence in the contagia afloat this year, liberties in the preparation of the infusions or defects in the construction of closed chambers which would have been of no moment a year ago were sufficient to ruin the experiments, and render nugatory the usual means of sterilization. Against such defects I continued to struggle. With a view to stopping all chinks and crannies which might permit of the entrance of contamination, I had some of the chambers carefully coated with oil-silk and others covered with three coatings of strong paint; and as failure had attended my efforts to procure an uninfected atmosphere upstairs, I had the entire apparatus used for digesting, filtering, and boiling removed to a store-room at the base of the Royal Institution. The floor of the room was of stone, and it was covered by no carpet. Prior to going into it, moreover, I caused my assistant to remove the clothes which he had previously

worn in the laboratory and to dress himself in others. The infusions prepared under these conditions were cucumber, melon, turnip, and artichoke, which, from beginning to end, were operated on below stairs. Two chambers were devoted to each infusion, and after the usual boiling in the chambers they were permitted to remain in the store-room throughout the night, being transferred to the warm laboratory next morning.

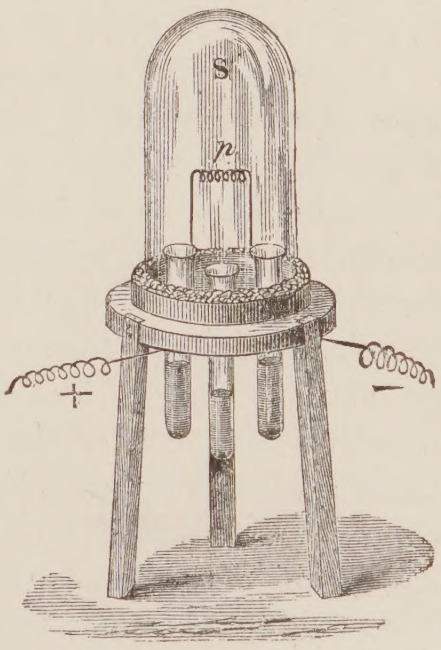
I fully expected that the majority of these chambers would prove sterile. I did not expect to find them all in this condition, because the chambers had been put together in the laboratory, the air of which must have deposited its germs not only on the glycerine-coated interior of the chambers, but also on the inner surfaces of the test-tubes. My expectation, moderate as it was, was not realized. The only noticeable peculiarity in the deportment of the infusions was that they yielded tardily, but in the end every one of them, without exception, broke down.

Was the infection in this case derived from the air of the store-room? I think not; and for this reason:— On the 27th of December four hermetically-sealed flasks, charged with a cucumber-infusion which had remained perfectly pellucid for some weeks, were opened in the store-room; four similar flasks, charged with the same infusion, were opened at the same time in the laboratory. On the 31st of December the whole group of the latter four was found invaded by organisms, while those opened in the store-room contracted no infection and developed no life. I do not imagine, therefore, that the air of the store-room had anything to do with the contamination of the infusions contained in the closed chambers, but conclude that the contagium already existed in the chambers when they were taken down stairs. They acted as infected houses placed in salubrious air.

§ 12. *Infusions protected by Glass Shades containing Calcined Air.*

I have already described this mode of experiment.[1] The shades stood upon circular plates of wood, each supported on a tripod (see fig. 16). Under each shade were two upright rods of stout copper wire, and stretching from rod to rod was a spiral (p) of platinum wire. The copper wires passed through the slab of wood, their free ends being in the air. The rim of each shade was surrounded by a collar of tin attached by wax to the slab, with a space of about half an inch between the collar and the glass. After the introduction of the infusions and the mounting of the shades, this annular space was packed with cotton-wool. The aim here was to destroy the floating matter of the air by the incandescent platinum spiral. The air heated by the spiral would of course expand, passing outwards through the cotton-wool, while the air re-entering, on the cooling of the shade, would be duly sifted by the wool. In my former experiments five minutes' incandescence sufficed to render the air absolutely inoperative on infusions exposed to its action.

FIG. 16.



In the present experiments the period of incan-

[1] Phil. Trans. vol. clxvi. p. 50.

descence was doubled, ten minutes being allowed instead of five, while the wire was raised to the highest possible degree of incandescence. The infusions employed were turnip and cucumber, a group of three tubes being charged with each. After the air had been calcined, the infusions were boiled for five minutes in an oil-bath. With this mode of treatment not a single failure occurred in 1875, turnip-infusion being among the number of liquids thus treated. This year two days sufficed to render every one of the six tubes turbid with organisms and to cover the infusion with a heavy scum.

I, however, had occasion to doubt the closeness of these shades. The wax intended to seal the junction of the tin collar with the plate of wood had cracked and yielded here and there, and the entry of contamination through such cracks was possible. Six new shades were therefore mounted and surrounded by collars which were imbedded in white lead and firmly screwed down to the plate of wood. The height of the collar, which measured the depth of the filtering layer of wool, was much greater than it had ever been last year. As before, the period of incandescence was ten minutes, during which the platinum spiral was brought as close as possible to its point of fusion.

Each of these six shades covered a group of three test-tubes. Two such groups were charged with turnip, two with cucumber, and two with artichoke-infusion. The infusions, as usual, were boiled for five minutes after calcination. They were all brilliant when prepared; but in two days every one of them had become turbid, and had covered itself with a fatty scum. This gradually augmented until it reached in some of the tubes a thickness of half an inch. The weight of the scum caused it in some cases to bag downwards, forming a kind of inverted cone, the apex of which was

more than an inch from its base. These bags finally broke and scattered their organisms in the subjacent liquid. Thus the conflict went on.

§ 13. *Further precautions against Infection.*

At the beginning of December, my attention being keenly aroused by those successive failures, I watched more closely than I had previously done the filling of the test-tubes through the pipette. Now and then I noticed minute bubbles of air carried down with the descending infusion. On escaping from the end of the pipette, these small bubbles, I concluded, would break, and scatter such germs as they contained in the air of the chamber. Last year I should have found it difficult to believe that a cause so small could lie at the root of the observed anomalies; but this year I had learned to respect small causes, and accordingly took measures to effectually exclude the air.

On December 4th three chambers, which had been previously left quiet for several days, were charged with carefully prepared cucumber-infusion, and two other chambers with turnip-infusion prepared with equal care. The following precautions were taken:—The funnel of the pipette formerly employed was broken off from its shank, and for it was substituted a 'separation-funnel' with a glass stopcock. This was connected by closely fitting india-rubber tubing with the shank of the pipette. But before the connexion was made, the funnel was filled with the infusion, and the stopcock turned on for a moment, until the liquid issued from the orifice below. The stopcock being then turned off, the flow of the liquid ceased, and the column in the shank below the stopcock was supported by atmospheric pressure. A pinchcock nipped the india-rubber tube at

its centre. The portion of tubing above the pinchcock was then filled with the infusion, and the end of the separation-funnel introduced into the tube, all air being thus excluded. On turning on the stopcock and releasing the pinchcock, the liquid passed slowly down the shank of the pipette, filling it wholly. The point of the shank was then placed in succession over the test-tubes, the infusion entering them without a single associated bubble. The arrangement was not absolutely perfect, but it was an improvement upon previous ones. As before, the charged chambers were placed in a room the air of which was maintained at a temperature of about 90° Fahr.

The result was as follows:—Of the two turnip chambers prepared as just described, on the 4th, one had completely given way on the 6th. In the other chamber two out of the three tubes had given way, but the third remained permanently brilliant. Previous to this series of experiments I had never succeeded in saving even a single tube of cucumber-infusion; here, however, two out of the three chambers charged with it remained perfectly clear for many days. Subsequently one of these chambers yielded in part, through an accident, but the other chamber is as brilliant at this moment as it was on the day of its preparation many months ago.

Now, as regards inherent power to generate life, the infusion of this chamber was in precisely the same condition as its two neighbours. They, one and all, contained the same infusion; and there is no way of accounting for the observed difference of deportment save by reference to contamination from without. Here we are certainly on the trace of the enemy which has given us so much trouble.

On the 5th of December two additional cases were

charged with infusion of melon prepared in the usual way; and on the 12th of December I subjected both these chambers, and those prepared upon the 4th, to a very close scrutiny. The result was instructive. After the introduction of the infusions, and prior to the removal of the separation-funnel, the india-rubber tubing connecting the latter with the shank of the pipette was perfectly closed by the pinchcock. Provided the clasping of the india-rubber tube round the shank of the pipette were perfectly air-tight, the liquid contained in the shank ought to remain there, supported by atmospheric pressure. If, however, the india-rubber tube failed to clasp with sufficient tightness the pipette-shank, air would insinuate itself between the two, and the depression of the liquid would be the consequence. The result observed upon the 12th was this:—In two only of the seven chambers prepared on the 4th and 5th was the liquid column found perfectly supported; and only in these two chambers were the test-tubes, which contained cucumber-infusion, without exception pellucid.

In the five remaining chambers the liquid columns, which had completely filled the pipette-shanks on the 4th and 5th, were found more or less depressed. The tubes in one of the chambers, containing melon-infusion, had become rapidly turbid and covered with scum. The pipette-shank in this case was found entirely emptied of its liquid and filled with air. Another chamber had nine inches, while a third had seven inches of its pipette-shank filled with air. In a fourth chamber only one inch of the pipette-shank was filled with air; here one out of a total of three tubes remained pellucid. Thus, where the closure above was perfect, we had in this instance perfectly pellucid infusions; where it was grossly defective, the infusions gave way in all the tubes;

while where the closure was but slightly defective we had the escape of a fraction.

The defects thus revealed came, I concluded, into play when the infusions were introduced, the descending column of liquid sucking in minute air-bubbles between the india-rubber tubing and the pipette, thus carrying with it the external contagium. Few seem aware of the precautions which are sometimes essential to save the experimenter from error in inquiries of this nature. Even with some of our best and most celebrated observers I find no adequate sense of the danger involved in their modes of experimentation.

§ 14. *Experiments in the Royal Gardens, Kew.*

But it was only in exceptional instances, dependent on the state of the air, that even precautions such as those described in the foregoing section secured freedom from contamination. The contagium seemed omnipresent and persistent, and whether it was local or general—due to the accidental condition of our laboratory, or to an epidemic of the air—became a question with me, not by any means to be decided offhand. On this point, then, I held judgment in suspense. The infection was, to all appearance, fully accounted for by reference to the conditions under which I worked; but as regards outbreaks of epidemics the autumn had been a remarkable one, and it seemed well worth investigating whether it was not also a period prolific generally in the germs of putrefaction.

I resolved therefore to break away wholly from the Royal Institution, and, thanks to the friendly permission of Sir Joseph Hooker, I was enabled to transfer my apparatus to Kew Gardens. By the enlightened mu-

nificence of Mr. Jodrell, a new and very complete laboratory had been just erected there, and in it I sought a purer air than I could find at home.

My chambers hitherto had been constructed of wood, but those to be tested at Kew were made of block-tin, and they were carried direct from the tinman's to the gardens without being permitted to come near the infected air of Albemarle Street. At Kew the test-tubes employed were first cleansed with carbolic acid, then washed with a solution of caustic potash, afterwards swept out with distilled water, and finally raised almost to the temperature of redness by a Bunsen-flame. They were then fitted air-tight into the chambers with white-lead and tow.

The chambers were closed on the 3rd of January, and allowed to remain quiet until the 8th, when the two most refractory liquids that I had encountered in the laboratory of the Royal Institution were introduced into them. These were infusions of cucumber and melon. There were two chambers devoted to each infusion—four in all; and each chamber embraced three large test-tubes. The period of boiling was that found effectual last year, *i.e.* five minutes. The temperature of the room in which the chambers were placed was maintained, partly by hot-water pipes and partly by a gas-stove, at about 90° Fahr.—a temperature which had been proved eminently favourable to the development of *Bacteria*.

Tubes containing the same infusions were at the same time exposed to the common air of the Jodrell laboratory. These became rapidly turbid and covered with scum. My interest and anxiety during the early days of the trial of the protected tubes may be imagined. After eleven days' exposure they showed no signs of giving way. On the 19th of January the four chambers

were removed in a van from Kew, and shown in the evening of that day to the members of the Royal Institution, including many eminent Fellows of the Royal Society. The infusions were one and all brilliant, no trace either of turbidity or scum being found associated with any of them. During all my previous efforts (and they had been very numerous) I had never succeeded in saving a single tube of melon-infusion; here, however, every tube of both chambers was intact. The epidemic was thus localized, the obvious cause of it being the contaminated air of our laboratory.

A couple of days subsequent to the removal of the chambers from Kew, a single tube of the cucumber-infusion became turbid, its two neighbours in the same chamber remaining intact. Not one of the other tubes, either of melon or cucumber, gave way. They all remained as pellucid in London as they had been at Kew. Their removal from Albemarle Street to the city last year ruined many of our sterilized chambers. I was not therefore prepared to see so little damage done by the transport from Kew.

It may be remarked, in passing, that this infection of an infusion by mere mechanical shaking is an obvious proof that the contagium is not a gas or vapour, but that it consists of particles capable of being detached from the interior surface of the chamber, and endowed with the power of passing into active life.

Two other chambers were exposed at the same time in the Jodrell laboratory, the one containing beef- and the other sole-infusion. They are by no means so sensitive as the cucumber and melon, still one of the three beef-tubes broke down, becoming thickly turbid throughout. Right and left of this tube its two companions remained perfectly transparent. As an illustration of the externality of the contagium, the result was

more conclusive than it would have been had all three tubes remained intact; for had the power of developing the organisms which produced the turbidity been inherent in the infusions, its action would not have been confined to a single tube.

It will be understood that when the chamber is lifted from the oil-bath in which its infusions are boiled, the air within the chamber contracts, and an indraught is the consequence. If the entering air be properly sifted, by passing it through cotton-wool plugs, no harm is done; but if it enter an aperture unsifted, it carries its motes along with it. In the beef-chamber just referred to an aperture of this kind, about the size of a pin-hole, was detected. This obviously was the door through which the contagium entered. Through a similar but graver defect in its chamber the sole-infusion also broke down; but in a subsequent experiment with sole-infusion in the Jodrell laboratory, two-thirds of the whole number of tubes charged with it remained free from all trace of life.

§ 15. *Experiments on the Roof of the Royal Institution.*

With a view to making, nearer home, experiments similar to those made at Kew, I had a wooden shed erected on the roof of our laboratory. The shed was provided with benches, water and gas-pipes, and a stove for heating. To an infusion of cucumber, which I had found extremely intractable in the laboratory, my attention was first directed. Two tin chambers of three tubes each were prepared, and transferred to the shed from the workshop where they were made, without

being permitted to enter our laboratory. The cucumber used for the infusion was also kept clear of the infected air; it was sliced and digested in the shed, the infusion was there filtered, introduced into the tin chambers, and boiled subsequently for five minutes.

The result was not that expected. Not a single tube of either of these two chambers escaped contamination. They one and all behaved like the same infusion in the infected laboratory, becoming in three days turbid throughout and laden with fatty scum.

I have been daily and hourly impressed with the parallelism between these phenomena of putrefaction and those of infectious disease. A further illustration of this parallelism is here presented to us. The clothes of my assistants who prepared the infusion in the shed had been worn in the laboratory, a transfer of infection by one of the modes of transport known to every physician being the result. The thoughtful physician cannot indeed fail to see the absolute identity of deportment between the contagia with which he is familiar, and those assailants of my infusions against which I have been contending so long.

With regard to the shed my first step, after this preliminary failure, was to disinfect it. This was done by washing every part of it, first with a mixture of carbolic acid and water, and secondly with a solution of caustic potash. When the whole was well dried, new tin chambers furnished with new tubes were introduced. Cucumbers and beef fresh from the market were also digested in the shed, my assistant taking care to cover his legs with clean linen trowsers, and his body with a new blouse. There was one chamber devoted to the cucumber and another to the beef. Into the former the infusion was introduced on the 19th, and into the

latter on the 20th of March; each infusion was boiled for five minutes after its introduction.

Let us compare results and draw conclusions. At a distance of eight yards from the shed, viz. in the laboratory, infusions both of beef and cucumber refused to be sterilized by three hours' boiling. Indeed I have samples of both infusions which have borne five hours' boiling and developed multitudinous life afterwards. But the upshot of this experiment in the disinfected shed is, that every tube of the two chambers, though boiled for only five minutes, contains an infusion which, at the present hour, is as limpid as the purest distilled water.

What shall we say, then? is the infusion in the laboratory endowed with a generative force denied to the same infusion in the shed? Irrespective of the condition of the air, can a linear space of eight yards produce so remarkable a difference? It is only the confusion of mind still prevalent in relation to this subject that renders such a question necessary. Let me add that it suffices simply to wave a bunch of hay in the air of the shed to make it as infective as the laboratory air. Even the unprotected head of my assistant when his body was carefully covered sufficed in some cases to carry the infection.

If anything were needed to illustrate the extraordinary care necessary on the part of physicians and surgeons, both as regards the clothes they wear and the instruments they use, such illustrations are copiously furnished by the facts brought to light in this inquiry.

§ 16. *Preliminary Experiments on the Resistance-limit of Germs to the temperature of Boiling Water.*

While continuing the conflict and experiencing the defeats recorded in the foregoing pages, a remark of Professor Lister's sometimes occurred to me. To apply the antiseptic treatment with success, the surgeon must, he holds, be interpenetrated with the conviction that the germ theory of putrefaction is true. He must not permit occasional failures to produce scepticism, but, on the contrary, must probe his failures, in the belief that his manipulation, and not the germ-theory, is at fault. This may look like operating under a prejudice; but Professor Lister's maxim is nevertheless consistent with sound philosophy and good sense; and if I permitted a bias to influence me in this inquiry, it was one fairly founded on antecedent knowledge, which led me to conclude that the long line of failures above referred to would eventually be traced to my ignorance of the conditions whereby perfect freedom from contamination was to be secured.

I laboured to discover these conditions, and to learn something more regarding the nature of the contamination—its origin, persistence, and manner of action. When these researches began, five minutes' boiling, as I have frequently stated, sufficed to sterilize the most diversified infusions. Here we have frequently extended the time of boiling to ten and fifteen minutes, and, in some cases glanced at above, to immensely longer periods, without producing this result. I desired more exact knowledge as to the limit of endurance, and with this view, on the 22nd of December, had six 'pipette bulbs' charged with an infusion of cucumber, sp. gr. 1004. They were then plugged with cotton-wool, hermetically sealed, and subjected to the boiling temperature for 10

minutes. Six other bulbs, charged with the same infusion and treated in the same way, were boiled for 30 minutes. Finally eight bulbs, similarly charged, were boiled for 120 minutes.

On the 23rd of December three of the first group of bulbs, three of the second, and five of the third, had their sealed ends filed off, and were afterwards exposed to a tolerably constant temperature of about 90° Fahr. Not one of these twenty bulbs preserved itself free from life. On the 25th of December every one of them had given way to cloudiness and turbidity.

There was, however, a marked difference between the sealed and the unsealed bulbs. To the latter, it will be remembered, the air had access through the plug of cotton-wool, while to the former no air had access, save the small quantity imprisoned above the infusion when the necks of the bulbs were sealed. The aerated bulbs grew rapidly and thickly turbid, while a passing cloudiness was all that showed itself in the sealed ones. This soon disappeared, and left the infusions apparently intact. In fact it required some attention to detect the appearance of this fugitive life, which existed only so long as there was oxygen to sustain it. I have ranged the sealed and unsealed tubes side by side in groups. To the most cursory observation the difference between them is obvious. The experiment strikingly illustrates the dependence of the special organisms here implicated on the oxygen of the air.

The experiments were pushed still further on the 28th of December. Two bulbs of cucumber, two of melon, two of turnip, and two of artichoke were then plugged, sealed, and maintained at the boiling temperature for four hours. Six of the eight bulbs burst in the operation, but two of them, a bulb of melon and

one of cucumber, bore the ordeal uninjured. After cooling, their sealed ends being broken off, they were placed in the warm room. The melon remained permanently sterile, but in two days the cucumber-infusion became turbid and laden with fatty scum.

Eight similar bulbs were boiled on the same day for five hours and a half. Four of them burst, but four remained intact. Of these, two contained cucumber-, one melon-, and one turnip-infusion. Three out of the four bulbs were sterilized by the long-continued boiling, but one cucumber-bulb passed through the ordeal unscathed. Two days after the operation it swarmed with life, and was covered with a fatty scum formed of matted *Bacteria*.

Many similar experiments were subsequently made. On the 27th of January, for example, six bulbs of turnip infusion were boiled for 220 minutes, six for 300 minutes, and two for 305 minutes. Suspended in the air above each infusion was a sprig of old Colchester hay, this being purposely introduced to augment the chance of infection. Notwithstanding its presence the bulbs were one and all permanently sterilized. The specific gravity of the infusion was in all cases 1007.

The sprigs of old hay were afterwards shaken into the liquid, but they produced no effect. For weeks afterwards the infusion remained clear. Was this impotence to generate life due to the fact that the nutritive power of the infusion had been destroyed by the 'blighting influence of heat'? Not so; for when the same infusion was infected by a sprig of fresh hay, by a small pellet of cotton-wool rubbed against the dusty shelves of the warm room, or by a speck of another infusion containing *Bacteria*, it never failed to develop life. The only observed difference between the effect produced by the dry hay or dust and the living *Bacteria*

was purely a difference of time. Inoculation with the finished organisms acted more rapidly than infection with the dust, but the effects were the same in the end.

On the 27th of January also nine melon-bulbs were treated exactly like the turnip, being furnished with sprigs of old Colchester hay, plugged with cotton-wool, and hermetically sealed above the plugs. Six of them were boiled for 215 minutes, and three for 220 minutes. They were one and all permanently sterilized; but, like the turnip, all of them were open to infection by fresh hay, dry dust, or living *Bacteria*. The specific gravity of the melon-infusion was 1008.

§ 17. *Further Experiments on the Resistance-limit of Germs to the Boiling Temperature.*

The amount of boiling which turnip-infusion failed to withstand is shown by some of the foregoing experiments; but to determine the limit of its resistance we must begin with shorter periods. On the 1st of March, therefore, eight groups of pipette-bulbs were charged with turnip-infusion which had been prepared in an atmosphere purposely infected with the germs of old Heathfield hay. In every case, moreover, a sprig of the same hay was placed in the air above the infusion. The bulbs had their necks plugged with cotton-wool, and were hermetically sealed above the plug. In this condition the respective groups were boiled for the following times:—

1st	group	15 minutes.
2nd	,,	30 ,,
3rd	,,	45 ,,
4th	,,	60 ,,
5th	,,	75 ,,
6th	,,	90 ,,
7th	,,	105 ,,
8th	,,	120 ,,

After boiling they were removed, permitted to cool, had their necks broken off by a file, and were afterwards exposed to the temperature of our warm room. It may be remarked that the infusion gradually deepened in colour from the 15-minute period, where the colouring was hardly sensible, to the 2-hour period, where the colouring became deep yellow. The effect was doubtless due to the oxidation of the infusion, which, notwithstanding the colour, was in all cases highly transparent.

Two days after their preparation, every tube of the series had become turbid and had begun to cover itself with scum.

On March the 6th the periods of boiling were prolonged with a fresh infusion. Two groups of tubes were, on that day, exposed to boiling water for the following times:—

1st group	180 minutes.
2nd „	240 „

On the 8th of March all the members of the first group were turbid and covered with scum. The second group was completely sterilized. This latter result is quite in accordance with the experiments made on the 27th of January. Turnip-infusion was then boiled for periods varying from 220 minutes to 305 minutes, complete sterilization being in all cases the consequence. These results were subsequently checked by a continuous series of experiments extending over periods of boiling varying from one to six hours. Up to three hours the infusion resisted sterilization; but when the periods of boiling were prolonged to four, five, and six hours respectively, all the bulbs became permanently barren. The liquid continued in the highest degree transparent, and in colour a brilliant orange-brown.

Experiments intended to determine the limit of resistance of cucumber-infusion were made on the 24th of February. Nine pipette-bulbs were then charged, plugged, hermetically sealed, and subjected to the boiling temperature for the following times:—

1st bulb	. . . . .	15 minutes.
2nd „	. . . . .	30 „
3rd „	. . . . .	45 „
4th „	. . . . .	60 „
5th „	. . . . .	120 „
6th „	. . . . .	180 „
7th „	. . . . .	240 „
8th „	. . . . .	300 „
9th „	. . . . .	360 „

After boiling and cooling they had as usual their ends broken off by a file. The result here was that at the 5th bulb, which corresponded to a boiling for two hours, the life-development suddenly ceased. All the tubes boiled from three to six hours inclusive were completely sterilized.

The infusion in this case had been diluted by an accident, so that its specific gravity was not much above that of distilled water. On the 28th of February, therefore, a fresh infusion having a specific gravity of 1006 was prepared, and introduced into a series of bulbs exactly as in last experiment. The bulbs were exposed to the boiling temperature for the following times:—

1st bulb	. . . . .	15 minutes.
2nd „	. . . . .	30 „
3rd „	. . . . .	45 „
4th „	. . . . .	60 „
5th „	. . . . .	120 „
6th „	. . . . .	180 „
7th „	. . . . .	240 „
8th „	. . . . .	300 „
9th „	. . . . .	360 „

The result here was that at the 6th bulb, which corresponded to three hours' boiling, the life-development suddenly ceased. All the bulbs boiled from 15 minutes to 180 minutes inclusive proved fruitful; while from 240 minutes to 360 inclusive all were completely sterilized. As in the case of the turnip-infusion, the cucumber subjected to long periods of boiling assumed an orange-brown tinge.

Comparing these results with those obtained with the turnip-infusion, it will be observed that cucumber and turnip exhibit about the same resistant power: three hours' boiling, and less, failed to sterilize both of them; four hours' boiling, and more, rendered both of them permanently barren.

The cucumber-infusions prepared on the 22nd and 28th of February were connected with the atmosphere through the cotton-wool plugs; but no attempt had been made to remove its floating matter from the air above the infusions. On the 22nd, however, four bulbs of the infusion were also prepared, charged with filtered air, left unplugged, and hermetically sealed. The same was done with four bulbs on the 28th of February. Each group was subjected to periods of boiling of 15, 30, 45, and 60 minutes respectively. All of them became turbid; but it was interesting to notice the gradual and obvious fall of life from the 15-minute to the 60-minute period. Could the *Bacteria* have been counted, and the result graphically represented, the ordinate corresponding to the abscissa 15 would have been found very considerably longer than that corresponding to the abscissa 60.

The method of experiment here for the most part pursued was employed by Spallanzani and Needham. It was afterwards extensively applied by the late excellent

Professor Wyman, of Harvard College, while in 1874 it was materially refined and improved upon by Dr. William Roberts, of Manchester. The method is hampered by one grave doubt. The air, plus its floating matter, is imprisoned in the sealed bulbs, so that the heat applied has not only to destroy the germs clasped by the infusion, but also those diffused through the supernatant atmosphere. Now it is not certain whether an amount of heat which would be absolutely destructive to a germ embraced by a hot liquid may not be wholly ineffectual when acting on a germ floating in vapour or air. Throughout Spallanzani's and Needham's experiments, throughout those of Wyman and Roberts, and throughout my own, as reported in this section and the last, this possibility of error runs. Such experiments, in short, do not enable us to state with certainty the temperature at which an infusion is sterilized, because the germs which most pertinaciously oppose sterilization may not belong to the infusion at all, but to the adjacent air.

The most astonishing cases of resistance to sterilization observed by Wyman were associated with this particular mode of experiment. The possible action of the uncleansed air, moreover, was in his case augmented by the fact that he employed quantities of liquid, very small in comparison with the size of his flasks. In some of his earlier experiments the volume of air was more than thirty times that of the infusion. These relative volumes are represented in the annexed figure (fig. 17), copied from Wyman's Memoir of 1862.[1]

FIG. 17.

[1] Silliman's American Journal, vol. xxiv. p. 80.

§ 18. *Change of Apparatus. New Experiments with Filtered Air.*

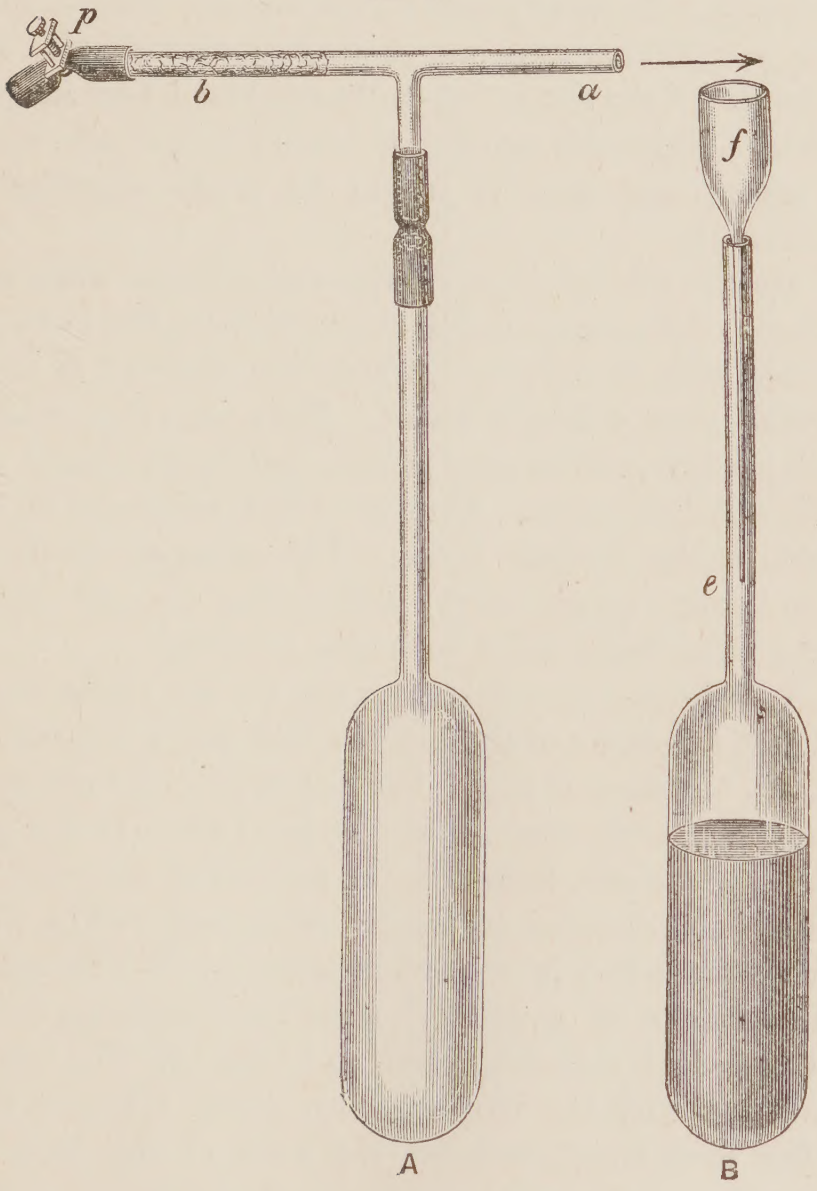
The source of possible error referred to in the last section had been long present to my mind, and I had already taken measures to avoid it. On the 2nd of January, 1877, an infusion of turnip (sp. gr. 1006) and an infusion of melon (sp. gr. 1008) were prepared and introduced into a series of pipette-bulbs in the following manner:—One end *a*, fig. 18, of a glass T-tube was connected with an air-pump, the other end *b* was closely plugged with cotton-wool, while to the third branch of the T-tube the neck of the pipette-bulb A was attached by india-rubber tubing. A piece of the same tubing, furnished with a pinchcock *p*, was also attached to the free end of the T-tube beyond the cotton-wool.

The bulb A was exhausted three times in succession, the pinchcock *p* being closed, and was three times filled with filtered air, the pinchcock being opened. At the third exhaustion the bulb was raised to a very high temperature by a Bunsen flame, and finally filled with filtered air. It was then plunged for a minute into ice-cold water, from which it was afterwards removed, detached from the T-tube, and then charged with the infusion by means of a narrow pipette, *f e*, shown at the top of B, fig. 18.

The rationale of the above proceeding is this:—On quitting the ice-cold water for the warmer air of the laboratory, expansion of the air within the bulb would occur. This would cause a gentle motion from within outwards, opposing all indraught of contaminated air. The entry of the infusion into the bulb would, I thought, also promote this outward motion. On the removal of the pipette, which occupied but a very small portion of the

neck of the bulb, a little warmth was applied to the latter, and during its application the neck was plugged with cotton-wool. The air entering through this plug to supply the place of the small quantity displaced by

FIG. 18.



the warmth would, I concluded, reach the interior of the bulb perfectly sifted of its floating matter. The necks of the bulbs were hermetically sealed, and the infusions

maintained for ten minutes at the temperature of boiling water. After a lapse of twelve hours their sealed ends were broken off by means of a file.

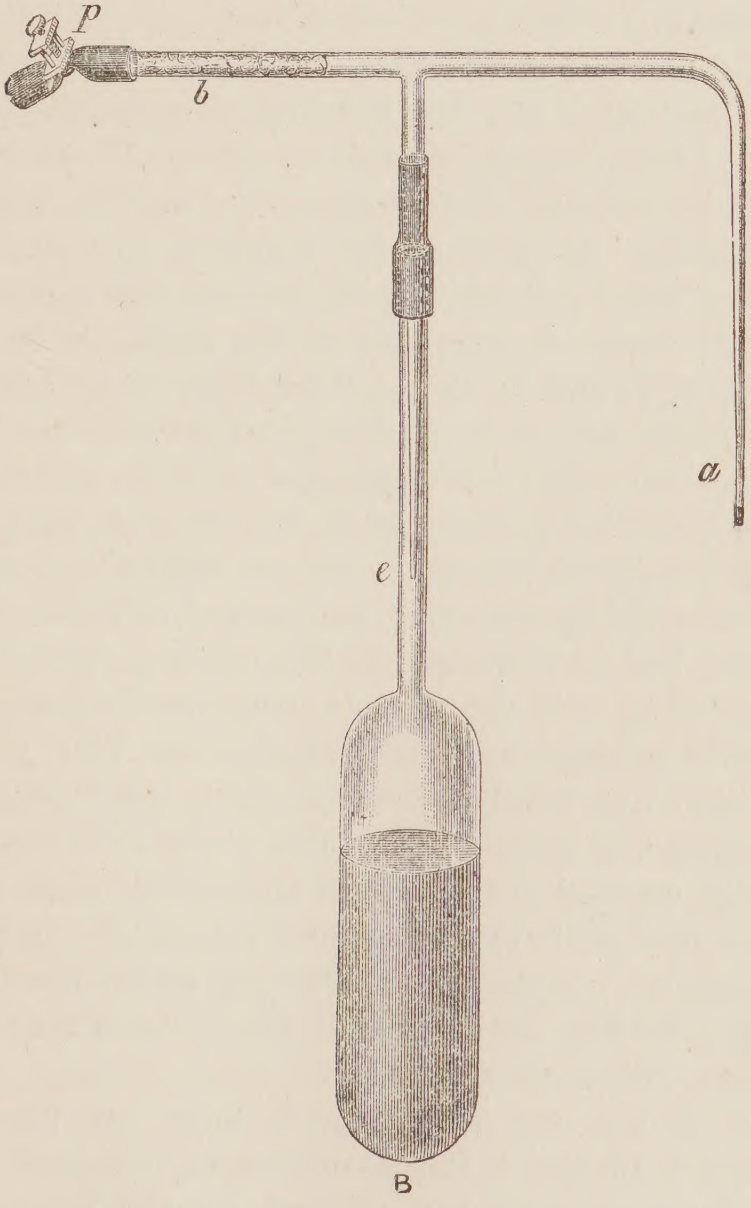
In our experiments on the 28th of December turnip and melon subjected to ten minutes' boiling entirely gave way. In these present experiments, where greater care was taken, two out of the six turnip-bulbs and three out of the six melon-bulbs remained permanently barren. Even this amount of success proved afterwards so exceptional that it might be fairly regarded as accidental.

On the 4th of January the experiments were continued. The pipette-bulbs employed were first carefully washed with carbolic acid, which was removed as far as possible with ordinary water. They were then washed with a solution of caustic potash, and finally rinsed out with distilled water. They were not subjected to the action of the Bunsen flame. The infusions employed were turnip (sp. gr. 1006) and melon (sp. gr. 1008), eight bulbs being filled with each infusion.

I could not be certain that the motion of the liquid fillet at the end *e* of the pipette with which the bulb B, fig. 18, had been charged, had not drawn into the neck of the bulb a modicum of the external air. In the present experiments, therefore, the method of charging the bulbs was modified in the following way:—The glass T-tube employed in our last experiments had its end *a*, which was to be connected with the air-pump, drawn out to a small orifice and bent as in fig. 19. The branch connected with the bulb was also drawn out to a tube of fine bore, which entered the neck of the bulb for some distance to *e*, the thicker part above being connected with the neck of the bulb by india-rubber tubing. The end *b*, as before, was plugged with cotton-wool and provided with a pinchcock, *p*. The object

here aimed at was that the liquid should be discharged into the bulb far below the india-rubber connecting-piece, and that during the discharge it should pass only through filtered air.

FIG. 19.



Each bulb was exhausted in the manner already described, and refilled three times in succession. When last filled it was plunged for a minute or so into

iced water, with the view of rendering the air within the bulb denser than that without. The pinchcock *p* being closed, the whole apparatus was then detached from the air-pump. On being lifted from the iced water into the warmer air there was a gentle outflow of air from *a*.

The mode of charging the bulb was this:—The point *a* was well sunk into the infusion, and the associated bulb, B, was plunged into boiling water. There was an immediate outrush of air from *a* which bubbled through the liquid. As soon as the bubbling had relaxed a little, *a* being still submerged, the bulb was transferred to iced water. A shrinking of the warm air was the consequence, and through *a* the infusion was forced by atmospheric pressure. It descended the middle branch of the T-tube, and was discharged from its end *e* into the bulb. The quantity of liquid obtained by a first immersion in the iced water was not sufficient to charge the bulb; but by repeating the process of heating and chilling two or three times, the point *a* never being permitted to quit the infusion, any required quantity was with ease and accuracy introduced. The neck of the bulb was finally detached from the T-piece by loosening the india-rubber tube. The bulb was then slightly warmed so as to cause an outflow of air from within, and while this outflow continued the neck was plugged with cotton-wool. It was sealed above the plug, and after the cooling of the infusion the sealed end was broken off with a file.

It is not my intention to take up time in describing in detail the numerous experiments made in accordance with this method, or the variety of infusions employed in testing its efficacy. Suffice it to say that, notwithstanding all my care, the results were chequered throughout. Sometimes success

would seem complete, but a repetition of the experiments—and I never felt safe without frequent and varied repetition—would, as before, present the success in the light of an accident. I am, however, secure in stating that while pursuing this plan I have in some cases effected complete sterilization by an amount of boiling which, in other cases, though twenty times multiplied, has failed to produce this effect. I have, for example, placed side by side in my collection two series of organic infusions, one as pellucid as distilled water, having been rendered permanently sterile by an exposure to the boiling temperature for five and ten minutes, and a second series containing the same infusions boiled for 30, 120, and 330 minutes respectively, and which nevertheless are muddy throughout and covered with scum. Even here, however, causes, other than differences of manipulation, may have contributed to the result.

Weeks of labour have been devoted to these experiments, nor did they exhaust the trials actually made. Another mode of proceeding was this. Pipette-bulbs were prepared by having a portion of their necks drawn out to a tube of very fine bore. The open end being connected with an air-pump, the bulb was exhausted and filled with filtered air several times in succession. In the final experiment the bulb was charged with one-third of an atmosphere of cleansed air; and while this pressure was maintained by the air-pump the narrow tube was hermetically sealed. Each bulb was afterwards heated almost to redness in the flame of a Bunsen lamp. It was charged by inverting the bulb, dipping the sealed end into the infusion, and breaking it off underneath the surface. The liquid entered until the bulb was two-thirds filled, when the narrow tube was again sealed. A great number of experiments were

thus executed, the results of which distinctly favoured the conclusion, though they did not to my satisfaction prove it, that the resistant germs were not to be wholly ascribed to the air, but that they had survived in the liquid.

§ 19. *Final proof that the Resistant Germs are embraced by the Infusion. Examples of Resistance both in Acid and Neutral Liquids.*

We here face a question which greatly harassed me at the time to which I now refer. It was this:—Have the germs, which under the circumstances here described produced life, been really embraced by the infusion itself during the time of heating? The liquid, it will be remembered, had to pass through the neck of the pipette-bulb, and it could not descend from the neck into the bulb without leaving a film adherent to the internal surface of the neck. This film, I reflected, might dry in part by evaporation; and it might, in doing so, leave germs behind which would be very differently circumstanced from those in the liquid. To germs thus exposed, not to the heat of water, but to the possibly less effective heat of vapour and air, the observed life might I thought be due. Before closing definitely with the proposition that the surviving germs had actually been in the liquid, the possibility to which I have just referred had to be inexorably shut out.

The evil was to some extent mitigated by charging the bulb, not through its own neck, but through a narrow tube issuing at right angles to the neck. But even here a portion of the neck and of the higher interior surface of the bulb was trickled over by the infusion. The difficulty was finally met, and completely surmounted, by causing the lateral tube to issue from

the centre of the bulb itself, and forcing the infusion into the bulb by atmospheric pressure, until the surface of the liquid stood clearly above the lateral orifice. To this level the liquid rose without wetting any portion of the surface against which it did not permanently rest.

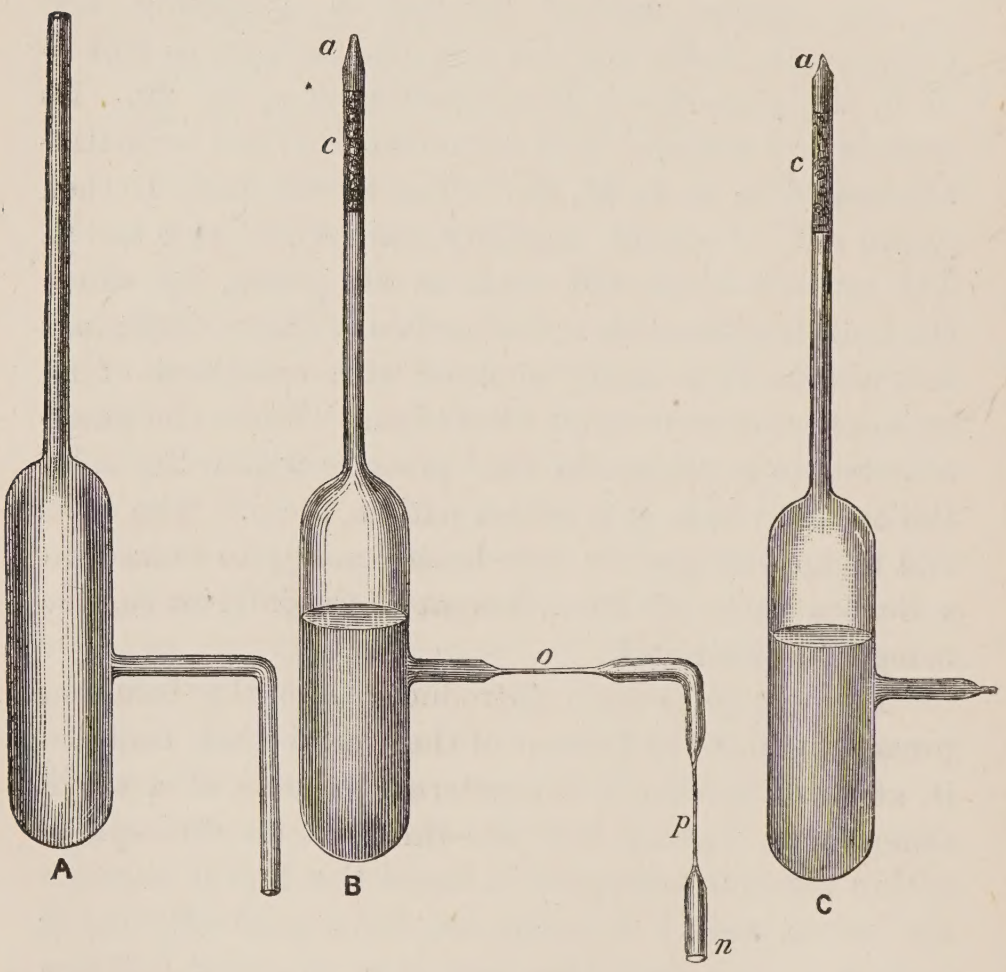
The precise method pursued in preparing and charging the bulbs was this:—First, the bulb as sent to us by the glass-blower is represented at A, fig. 20. Its neck is first plugged with cotton-wool (*c*) and hermetically sealed as at B, fig. 20. The lateral tube is then drawn out to almost capillary narrowness at *o* and *p*. The end *n* is connected with an air pump, by which the bulb is exhausted, and after two or three emptyings and fillings, it is finally charged with one-third of an atmosphere of thoroughly filtered air. While the pump attached to *n* maintains this pressure within the bulb, the capillary tube *p* is sealed with a lamp. The bulb and its appendages are then heated nearly to redness in a Bunsen flame, all life adherent to the interior surface being thus destroyed.

The end *p* is then introduced into the infusion, pressed against the bottom of the vessel that contains it, and thus broken. The external pressure of a whole atmosphere, having but one-third of an atmosphere within the bulb to oppose it, forces the liquid through the lateral tube. It enters the bulb, gradually rising until it reaches the orifice, and rises above it. When the pressure within is exactly equal to the pressure without, two-thirds of the bulb are occupied by the liquid.

The infusion then extends without breach of continuity from the bulb B to the vessel in which the end *p* is immersed, the uncleansed air being thus completely

excluded. A small gas flame is carefully applied at *o*. The liquid within the narrow tube vaporizes, and the vapour drives the liquid to some distance right and left from the place of heating. In the absence of the liquid the fine tube reddens, fuses, and is hermetically sealed.

Fig. 20.



The aspect of the bulb after it has been thus charged is shown at c, fig. 20.

By this method, on the 20th of February sixteen bulbs were charged with infusions of old Heathfield hay and of a hard wiry hay from Guildford, not old. They were divided into four groups, four bulbs in a group.

Each group embraced two acids and two neutral infusions. They were boiled for the following times:—

1st group	. . . . .	10 minutes.
2nd „	. . . . .	20 „
3rd „	. . . . .	30 „
4th „	. . . . .	60 „

After the bulbs had sufficiently cooled, their sealed ends were removed by a file.

On the 21st of February, less than 24 hours after their preparation, all these bulbs showed signs of yielding. On the 22nd they were all turbid, while as regards the comparison of acid and neutral infusions, their condition was this:—

1st group.	10 minutes.	Guildford neutral distinctly more turbid than Guildford acid. Scum on former, none on latter.
		Old Heathfield neutral not to be distinguished from old Heathfield acid. Both turbid and covered with scum. Much lightened in colour.
2nd group.	20 minutes.	Guildford neutral distinctly more turbid than the acid liquid.
		Old Heathfield neutral more turbid than the acid infusion.
3rd group.	30 minutes.	Guildford neutral a little more turbid than Guildford acid: difference small.
		Old Heathfield neutral more turbid than the acid infusion; the former with scum, the latter none.
4th group.	60 minutes.	Guildford neutral distinctly more turbid than Guildford acid; the former with scum, the latter free from scum.
		Old Heathfield neutral somewhat more turbid and scummy than the acid liquid: difference not great.

Here I take it to be absolutely certain that the germs which resisted sterilization were contained in the

liquid—a proof necessary to the argument, and admitting of no question, being thus offered to the reader.

On the 22nd of February four groups of bulbs were charged as above described with the same two infusions, and the periods of boiling prolonged as follows:—

1st	group	90 minutes.
2nd	„	120 „
3rd	„	180 „
4th	„	240 „

As in the last case, neutral and acid infusions of each kind of hay were operated on. This was the result:—On the evening of the 23rd, that is to say, 24 hours after their preparation, every one of these bulbs disclosed to the practised eye that organisms existed within it. At 2 p.m. on the 24th they all swarmed with life. The old Heathfield bulbs, both acid and neutral, were turbid and covered with scum, the very weakly-acid infusions being indistinguishable in appearance from the neutral ones. In the case of the Guildford infusion, however, the scum of the neutral infusions was richer and heavier than that on the acid ones.

Four hours mark the limit to which the boiling was carried in these experiments. On the 27th of February the periods of boiling were still further prolonged. Four groups of bulbs charged with infusions of the same two kinds of hay, both acid and neutral, were on that day boiled for the following times:—

1st	group	300 minutes.
2nd	„	360 „
3rd	„	420 „
4th	„	480 „

I had previously boiled infusions of old Heathfield, old London, and old Colchester hay for 5 hours, and

found them afterwards permanently barren. In the present instance, also, all the bulbs boiled for 5 hours, 6 hours, and 7 hours were completely sterilized. They remained ever afterwards perfectly brilliant. This, with one exception, was also the deportment of the group of bulbs boiled for 8 hours. The exception was a neutralized bulb of the Guildford infusion, which became turbid and covered with scum. Considering the severity with which the bulb had been treated prior to charging, and considering the mode of charging it, the life developed could not possibly have been the product of external germs. Through profound and thorough hardening and desiccation, through defect of contact with the liquid or some other cause, some germs in the infusion itself had, I doubt not, been enabled to withstand the extraordinary ordeal here described.

Was it 'the blighting influence of heat' that deprived the great majority of these 8-hour bulbs of the power of spontaneous generation? Whatever be the meaning attached to such language the reply is obvious, that the 'blighting' was the same for all the bulbs; and yet we find one of them which, when taken from the boiling water, was perfectly brilliant, rendered in two days muddy with organisms. Further, it was only necessary to wash with perfectly sterilized distilled water the adherent germs from a small bunch of hay, and to inoculate the clear infusion in an 8-hour bulb with the washing-water, to cause it within four-and-twenty hours to become turbid throughout. To speak more definitely, 14 hours in the warm room were found sufficient to cloud the infected 8-hour bulb with *Bacteria*. Thus the infusion, when living germs are restored to it, shows its perfect competence to develop them, and it was solely the destruction of the germs which it possessed before it was boiled that rendered it sterile afterwards.

It is worth bearing in mind that the particular kinds of hay whose germs manifested these extraordinary powers of resistance were so sapless and indurated that the specific gravity of their infusions, even after five hours' digesting, could not without difficulty be sensibly raised above that of distilled water. This was more especially the case with old Heathfield and old Colchester hay, the infusions of which, though highly coloured, were marked, almost without exception, 'specific gravity 1000.' Old London hay-infusion was usually 1003, while infusions of new Heathfield hay were raised without difficulty to 1007.[1]

As already stated, I never felt safe in these experiments until they were checked by careful repetition. A partial corroboration has been already adverted to. But on the 2nd of March I had the same infusions prepared, kept in a cool place throughout the night, and introduced into four groups of bulbs next morning. The infusions were all brilliant. During the process of boiling one group of bulbs blew up, hence one link is absent from the series. I thought the 4-hour group could be best spared, and therefore selected it for omission. We had thus the infusions subjected for one, two, three, five and six hours respectively to the boiling temperature. The three first hours agree exactly with their predecessors. No single bulb within these limits was sterilized, all of them became turbid throughout and loaded with scum.

[1] A comparative experiment on dried and undried peas, described in the Proceedings of the Royal Society (1877, vol. xxv. p. 503), may be referred to here as illustrating the manner in which desiccation restricts diffusion, and thus tends to preserve the integrity of the desiccated germ. I suppose the original mineral salts of the hay were still retained in the old samples; but hot water appeared to have little power of extracting them. The resistance of the hay in this respect appeared to be shared by its germs.

One bulb of the 5-hour group and one of the 6-hour group also became turbid and flocculent within, but without any scum upon the surface. As in the case of the 8-hour bulb already mentioned, this appearance of life was, I doubt not, due to stray germs of exceptional resisting power, which maintained themselves unscathed in the infusions after their fellows had been destroyed.

By multiplied experiments of a similar character executed subsequently, and fortified by others made in a different way, all doubts as to the real ordeal to which the germs had been exposed were set at rest. A flood of light, moreover, was thrown upon the difficulties recorded in the foregoing pages. Prior to the introduction into our laboratory of the particular samples of desiccated hay whose adherent germs had manifested such extraordinary powers of resistance, infusions of all kinds, even those of hay itself, were sterilized with ease and certainty. But the old London, the old Heathfield, the Guildford, and the old Colchester hay brought a plague into our atmosphere, and thus the infusions of other substances, some samples of hay included, became the victims of a pest entirely foreign to themselves. The failure to sterilize cucumber, turnip, beetroot, artichoke, melon, beef, mutton, haddock, herring, sole, was plainly due to the fact that their infusions had been prepared in an atmosphere, or brought into contact with vessels, contaminated with germs which have been here shown capable of resisting 240 minutes' boiling. It is obvious from all this that to speak of an infusion being rendered barren by such or such a temperature, is simply to use words without definite meaning; because the temperature at which any infusion is sterilized depends upon the character and condition of the germs which find access to it. The death-temperature, for

example, may be more than three hours in London, and less than three minutes at Kew.[1]

I will cite here two conspicuous illustrations of the infective energy of those desiccated hay-germs in two infusions which, under ordinary atmospheric conditions, are very easily sterilized. On the 30th of March five pipette-bulbs were charged with clear beef-infusion and boiled for the following times:—

1st bulb	. . . . .	60 minutes.
2nd „	. . . . .	120 „
3rd „	. . . . .	180 „
4th „	. . . . .	240 „
5th „	. . . . .	300 „

After cooling, the sealed ends were broken off, the air being admitted through cotton-wool plugs. Every one of these bulbs became charged with organisms. In the shed, eight yards off, this beef-infusion was, as already reported, sterilized by five minutes' boiling.

Precisely the same experiment was made on March 30 with pellucid mutton-infusion. Not one of the five bulbs was sterilized. All of them are at this moment charged with life. It behoves those engaged in the industry of preserved meat and vegetables to keep clear of the old-hay contamination. Probably they from time to time have encountered difficulties and disappointments which they could not explain, but which may be solved by reference to the results here set forth.

[1] I have already described the distribution of *Bacteria*-germs in the air as 'Bacterial clouds.' Were our vision sufficiently sharpened to see the manner in which such germs are distributed over the surface of a meadow, we should not, I am persuaded, find that distribution uniform. We should, in my opinion, find the germs grouped in crowds, with comparatively free interspaces, like violets on an alp, or mushrooms in a field. It is therefore conceivable that two bunches of hay from the same meadow may differ from each other in deportment.

But above all the practical question arises:—May not the surgeon have to fight sometimes against enemies like those here described? The particular germs with which I have been so long contending act on both fish and flesh in the manner described. How would they behave in the wards of an hospital? Can they cause wounds to putrefy? and if so, would they succumb to the disinfectants usually applied? These are questions the weighty import of which will be best understood by the enlightened follower of the antiseptic system, and which he will know how to answer for himself.

§ 20. *Remarks on Acid, Neutral, and Alkaline Infusions.*

In the foregoing section reference was made to the comparative deportment of acid and neutral infusions. There can be no doubt of the fact that, for the nutrition and multiplication of *Bacteria*, acid infusions are less suitable than neutral or slightly alkaline ones. In acid infusions exposure to the common air sometimes copiously developes *Penicillium*, while it fails to develope *Bacteria*. It is also true that exposure for a certain time to a certain temperature may in many cases prevent the appearance of life in an acid infusion, and fail to prevent it in a neutral or slightly alkaline one.

In the present inquiry this has been frequently found to be the case. I have many closed chambers, for example, to which the process of 'discontinuous heating,' to be subsequently described, has been applied; and with them it has proved a common experience that an amount of heating which has rendered acid infusions of hay permanently barren has failed to sterilize the corresponding neutral infusions. Moreover, in the

cases just recorded, where single bulbs escaped steriliz ation though exposed for five, six, and eight hours to the boiling temperature, it was always a neutral bulb that kindled into life. To these instances another may be added here. On the 22nd of March an infusion of the wiry Guildford hay already referred to was divided into two parts, one of which was neutralized and the other left acid. Five pipette-bulbs were filled with the one infusion and five with the other. After hermetic sealing they were all completely submerged in water and boiled for six hours. Every one of the acid bulbs was sterilized by this process, while in two days three of the five neutral ones became turbid and covered with scum.

The best thought that I have been able to bestow upon this subject does not induce me to lean towards the explanation suggested by M. Pasteur, namely, that the germs escape the destructive action of the heat because they are not wetted by the alkaline or neutral liquid. From the comparative action of alkalized and acidulated water upon hay, I should be inclined to infer that the wetting of its germs by the former would be more prompt than by the latter. The question, I think, is not one of wetting, but of relative nutritive power. Two *Bacteria*-germs of equal vital vigour dropping from the atmosphere, the one into a neutral or slightly alkaline, the other into an acid infusion, soon cease to be equal in vigour. The life of the one is *promoted*, the life of the other only *tolerated* by its environment. When the temperature surrounding both is raised to a prejudicial height the one will suffer more than the other, because equally inclement conditions are brought to bear upon constitutions of different strengths; and if the temperature be sufficiently exalted or sufficiently prolonged to become fatal, the more weakly organism

will be the first to give way. A germ, moreover, brought close to the death-point in a neutral or in an alkaline infusion may revive, while in an acid one it may perish—just as proper nutriment may rescue a dying man, while improper nutriment would fail to do so. These elementary considerations, founded on the demonstrable fact that *Bacteria*-germs are more fully vivified and better nourished in neutral infusions than in acid ones, suffice, I think, to explain the observed difference of action. At all events, these are the thoughts which have become rooted in my mind, through long observation and long pondering of this question.[1]

§ 21. *Remarks on the Germs of* Bacteria *as distinguished from* Bacteria *themselves.*

The failure to distinguish between these stubborn germs and the soft and sensitive organisms which spring from them has been a source of error in writings on Biogenesis. In his able and important paper, 'On the Origin and Distribution of *Bacteria* in Water, and the circumstances which determine their existence in the Tissues and Liquids of the Living Body,' Dr. Burdon Sanderson, for example, has described experiments from which, in my opinion, very incorrect conclusions have been drawn. He exposed to the common air vessels containing Pasteur's solution, which when inoculated with fully developed *Bacteria* enables them freely and copiously to increase and multiply;

[1] From their deportment in boiling, I should infer that the air dissolved in an alkaline liquid is in a different physical condition from that dissolved in an acid liquid; and to this, in some measure, the difference of nutritive power may be due. I have been unable to find any experiments on the comparative absorption of air by acid and neutral liquids. The subject is, I think, well deserving of attention.

he even caused the air to bubble through the solution, and finding that though *Torula* and *Penicillium* were luxuriantly developed in the liquid, *Bacteria* never made their appearance, he concluded, 'not merely that the conditions of origin and growth of *Bacteria* and fungi are considerably different, but that, as regards the former, the germinal matter from which they spring *does not exist in ordinary air*.'[1]

Dr. Sanderson subsequently reaffirmed the position here laid down. 'In my preceding experiments,' he says, 'it has been shown that, although *Torula*-cells and *Penicillium* appear invariably, and without exception, on all nutritive liquids of which the surfaces are exposed to the air, without reference to their mode of preparation, no amount of exposure has any effect in determining the evolution of *Bacteria*.'[2] And, again, with reference to another experiment:—'The result shows that ordinary air is entirely free from living *Bacteria*.'[3] His general conclusion is that, as regards the development of *Bacteria* in organic liquids, 'water is the contaminating agent.'

Upon these experiments, and the conclusion drawn from them, an argument has been founded by Dr. Bastian,[4] which would be weighty were its basis sure. In reference to the Presidential Address of the British Association in Liverpool,[5] he argues thus:—'Speaking of living *Bacteria*-germs, Professor Huxley summed up by saying, "considering their lightness, and the

[1] Appendix to the Thirteenth Report to the Medical Officer of the Privy Council for 1871, p. 335.

[2] 'Appendix,' p. 338. Though Dr. Sanderson speaks of 'all nutritive liquids,' if I understand him aright, he really tried but one, and that was a mineral solution, not an animal or vegetable infusion.

[3] 'Appendix,' p. 339.

[4] 'Evolution,' p. 44.

[5] Brit. Assoc. Report, 1870.

wide diffusion of the organisms which produce them, it is impossible to conceive that they should not be suspended in the atmosphere in myriads." Had Professor Huxley himself made some careful and discriminating experiments on this part of the subject, he might have found that the supposed impossibility of conception was entirely delusive. . . . . What has been the subsequent progress of events? In the first place, it has been shown by Professor Burdon Sanderson, myself, and others. that the living *Bacteria*-germs are not diffused through the air to any appreciable extent; and this is now a very widely accepted doctrine, in spite of its being, as Professor Huxley imagined, an impossible conception.' The 'others' referred to by Dr. Bastian embrace among them, it is to be admitted, the celebrated naturalist Cohn.

Dr. Bastian was quite correct in saying that the 'doctrine' he enunciated was, at that time, 'widely accepted.' But the deportment of almost any sterilized animal or vegetable infusion exposed to common air will disprove the doctrine. It is irreconcileable with the experiments on melon-, turnip-, cucumber-, and hay-infusions, alluded to in this memoir. Such infusions, after having been sterilized by exposure for six or eight hours to the boiling-temperature, remain, if protected from the *Bacteria*-germs of the air, for ever barren; but when infected spontaneously, or purposely, by atmospheric germs, they are found, within eight and forty hours after such infection, thickly crowded with *Bacteria*. That London air is laden with living *Bacteria*-germs is as certain as that London chimneys are laden with smoke. What Dr. Sanderson's extremely important experiments really prove is, that a mineral solution competent to nourish the *Bacteria*, after they have been fully developed, is not competent (or, rather,

is but very feebly competent) to effect the transfer from the germ state to that of the finished organism. It can feed the chick, but it cannot hatch the egg. As I have already expressed it, the experiment proves, not the absence of *Bacteria*-germs from the air, but the inability of the mineral solution to develop them.

Another experiment, described by Dr. Sanderson in the paper above referred to, is this:—'A glass rod was charged with *Bacteria* by dipping it into a solution on the surface of which there was a viscous scum, consisting entirely of these bodies imbedded in a gelatinous matrix. The rod was allowed to dry in the air for a few days. It was then introduced into boiled test-solution contained in a superheated glass. On February 6th the liquid was already milky, and teemed with *Bacteria*.

'To determine the effect of more complete desiccation, an éprouvette containing one cubic centimeter of cold water, previously ascertained to be zymotic, was evaporated to dryness in the incubator, and kept for some days at a temperature of 40° Cent. On February 20th the dried glass was charged with boiled and cooled solution, and plugged with cotton-wool in the usual way. The liquid was examined microscopically on March 2nd, when it contained numerous *Torula*-cells, but no trace of *Bacteria*. It therefore appears that the germinal particles of *Bacteria* are rendered inactive by drying without the application of heat.'

These experiments have been quoted as conclusive in reference to the influence of desiccation. They are quoted, moreover, as applicable not only to the developed *Bacterium*, but also, without restriction, to the germs from which *Bacteria* spring. 'To maintain,' says Dr. Bastian,[1] 'his Panspermism in the face

[1] 'Evolution,' p. 156.

of his own experiments, Spallanzani was compelled to assume that the germs of the lower infusoria do possess this seed-like property of developing after desiccation. Modern science, however, declares that they have no such property. We are told most unreservedly by Professor Burdon Sanderson, not only that the germinal particles of *Bacteria* are rendered inactive by thorough drying, without the application of heat, *i.e.* by mere exposure to air for two or three days at a temperature of 104° Fahr., but also that fully-formed *Bacteria* are deprived of their power of further development by thorough desiccation.' In this unqualified sense the conclusion is untenable. I could cite a multitude of experiments to prove this, but a reference to one or two of them will here suffice.

A small bunch of old Heathfield hay was washed with distilled water, which was received into a champagne-glass. The glass was placed on a stove until the water had all evaporated, and the dried residue was permitted to remain upon the stove for several days. Dr. Sanderson's drying temperature was 104° Fahr., mine was 120° Fahr., and my period of drying was longer than his. Scraping a little of the dry sediment treated as above from the bottom of the champagne-glass, I infected with it a bulb containing hay-infusion which had been completely sterilized by eight hours' boiling. When infected, the infusion was brightly transparent, but forty-eight hours afterwards it was teeming with *Bacteria*. With regard to the doctrine that these organisms arise from 'dead organic particles,' instead of from living germs,[1] no scientific man at the present day ought, I submit, to be called upon to spend a moment's thought upon it.

One other reference will suffice. I have had bun-

[1] 'Evolution,' p. 44.

dles of hay hung up for seven or eight weeks in the hot rooms of the Turkish Bath in Jermyn Street, and exposed during the whole of this time to a temperature of 140° Fahr. and upwards. The germs adherent to this hay were not killed by even this amount of desiccation. When a sterilized animal or vegetable infusion was infected with them they gave birth in the usual time to swarms of *Bacteria*.

§ 22. *Sterilization by discontinuous Heating.*

Keeping the distinction between germs and developed organisms here insisted on, and the probable changes that occur in passing from the one to the other, clearly in view, I have been able to sterilize with infallible certainty the most obstinate infusions referred to in this paper, without either raising the temperature of the infusions beyond their ordinary boiling-point, or inordinately prolonging the application of heat. The infusions may be sterilized by a temperature even below that of boiling water, while the time of its application may be but an extremely minute fraction of that resorted to in some of the foregoing experiments.

It is an undisputed fact that active *Bacteria* are killed by a temperature far below that of boiling water. It is also a fact that a certain period, which I have called the period of latency, is necessary to enable the hardy and resistant germ to pass into that organic condition in which it is so sensitive to heat. There can hardly be a doubt that the nearer the germ approaches the moment when it is to emerge as the finished organism, the more susceptible it is to that influence by which that organism is so readily destroyed. We may learn from experience, aided by the power of search

which the concentrated luminous beam places in our hands, what is the approximate time required for the Bacterial germ to pass into the *Bacterium*. Say that it is twenty-four hours. Supposing the heat of boiling water, or even a lower heat than that of boiling water, to be applied to the germ immediately before its final development, when all its parts are plastic, when it is, in short, on the point of reaching a stage at which a temperature of 140° Fahr. is demonstrably fatal. It is in the highest degree probable that a temperature of 212°, or of 200°, or, indeed, a temperature of 150°, if applied sufficiently often or for a sufficient length of time, will prove fatal to the germ, and prevent the appearance of the still more sensitive organism to which the germ is on the point of giving birth.

Here, at all events, we have a theoretic finger-post pointing out a course which experiment may profitably pursue. It is not to be expected that the germs with which our infusions are charged all reach their final development at the same moment. Some are drier and harder than others, and some, therefore, will be rendered plastic and sensitive to heat before others. Hence the following procedure.

Four-and-twenty small retorts were charged with hay-infusions on the 1st of February, and subjected morning and evening to the boiling temperature for one minute. The last heating took place on the evening of the 3rd of February. The retort-necks had been plugged with cotton-wool; the air within them, however, had not been filtered, and there was comparatively little care bestowed on their preparation. After the final heating they were abandoned to the temperature of a room kept close to 90° Fahr. A series of similar retorts charged at the same time with the same infusions were boiled continuously for ten minutes, plugged

while boiling, and placed in the same warm room. Two days after their preparation the retorts last mentioned had, without a single exception, given way to turbidity and scum. On the other hand, twelve of the twenty-four retorts which had been subjected for a much shorter period to the discontinuous boiling remained permanently brilliant and free from scum.

On the same 1st of February eight pipette-bulbs were charged with two hay-infusions, four bulbs being devoted to each. The air above the infusions was the unfiltered air of the laboratory. They were subjected to the temperature of boiling water for a minute; at the same time four other bulbs containing the same infusions were boiled continuously for ten minutes and suspended beside their neighbours. Twelve hours subsequent to their first brief heating the eight bulbs were perfectly brilliant, and while in this condition they were again subjected to the boiling temperature for a minute. On the evening of the same day they were subjected to the boiling temperature for half a minute, and on the following morning the process was repeated. Two additional heatings of the same brief character were resorted to. The result of the whole experiment was that two days after their preparation the four bulbs which had been boiled for ten minutes were found turbid and covered with scum, while two months after their preparation the eight bulbs whose periods of boiling added together amounted only to four minutes were perfectly brilliant and free from scum.

The reason of this procedure is plain. By the first brief application of heat the germs, which are at that moment plastic, are killed; and before any of the remaining germs can develop themselves into *Bacteria* they are subjected to another brief period of heating. This again kills such germs as are sufficiently near their

final development. At each subsequent period of heating the number of living germs is diminished, until finally they are completely destroyed. The infusion, if protected from external contamination, remains for ever afterwards unchanged, although, when living *Bacteria*, a sprig of hay, or even the dry dust particles of the laboratory are sown in it, the sterilized liquid shows its power both of enabling the fully developed organism to increase and multiply, and of developing the desiccated Bacterial germ into multitudinous Bacterial life.

On the same date an experiment was made with a series of pipette-bulbs, whose necks were so bent and plugged with cotton-wool that no impurity from the wool could fall into the infusion. Four bulbs were charged with an infusion of Heathfield and four with an infusion of London hay, samples of the same infusions being introduced at the same time into another series of bulbs which were plugged like their neighbours and subjected continuously to the boiling temperature for ten minutes. The eight bulbs first referred to were, on the contrary, discontinuously boiled, the sum of their periods of boiling being four minutes. The result is that while the entire series of bulbs boiled for ten miuutes gave way within forty-eight hours after their preparation, seven out of the eight bulbs which had been subjected to discontinuous boiling remained permanently brilliant.

On the 3rd of February, with the view of testing the new method still further, infusions of our most refractory kinds of hay were prepared. There were five bulbs of neutralized Guildford infusion, and five of a neutralized infusion formed from a mixture of old Colchester and old Heathfield hay. Two bulbs of each infusion were at the same time charged and subjected to the boiling temperature for ten minutes. The ten bulbs

first mentioned were never raised to the boiling temperature at all, the maximum to which they were exposed being some degrees below their boiling-point. The result is that while every one of the four bulbs boiled for ten minutes has become turbid and covered with scum, one only of the ten discontinuously heated bulbs has given way; nine of them remain as brilliant as at first.

It is obvious from what has gone before that two hundred and forty minutes might have been substituted for ten minutes without altering this result. Five minutes of discontinuous heating can accomplish more than five hours' continuous heating.

On the same date three bulbs charged with an acid infusion of London hay were subjected to the same discontinuous treatment. They all remain brilliant to the present hour.

On the 7th of February four of Cohn's tubes were charged with turnip-infusion, which was heated discontinuously night and morning up to a temperature of 205° Fahr. The total time during which they were exposed to this temperature was about three minutes. They were permanently sterilized, and exhibit a singular brilliancy to the present hour.

The discontinuous method of heating has also been applied with success to the closed chambers. One mode of operation is this:—An oil-bath is heated to a temperature of 300° Fahr. The charged test-tubes of the closed chamber are then plunged in the oil, which clasps the tubes to the level of the surface of the infusion. They are either raised to incipient boiling and then removed, or suffered actually to boil for thirty seconds and then removed. Another mode of heating is this: instead of being plunged into hot oil, the test-tubes are plunged for two or three minutes into boiling water,

taken out, wiped dry, the actual boiling being finished by a spirit-lamp. This is a very handy method, and more under the experimenter's control than the oil-bath. When the latter is employed, the infusions sometimes in great part waste themselves by leaping from their tubes; but the spirit-lamp enables us to humour the infusions by occasionally withdrawing the flame and moderating the ebullition. The lamp, of course, may be employed alone without the preliminary immersion of the tubes in hot water. Usually the process of heating is repeated at intervals of twelve hours, but in the case of very nutritive infusions in a very warm room the interval ought to be shorter. Practice must inform the experimenter on this point. The reheating must always occur before the infusions show the slightest visible tendency to change.

In the early days of February a closed chamber of six tubes was treated in the manner here described. Three of the tubes were charged with strong turnip- and the three others with strong artichoke-infusion. After two days' discontinuous heating night and morning, they were allowed to remain undisturbed in the warm room. The six tubes remain perfectly brilliant to the present hour.

On the 12th of February a closed chamber of three tubes was charged with cucumber-infusion. Heated discontinuously in the manner described, and abandoned afterwards to a warm temperature, the three tubes remained perfectly sterile.

Any process competent to sterilize very old hay can sterilize with greater ease any other infusion. The fact, therefore, that only a few days ago three closed chambers charged with our most refractory hay-infusions were sterilized by discontinuous heating proves the power of the method over infusions of all kinds.

By this method very instructive comparative experiments might be made, and the resistant power of different germs might be expressed in terms of the heatings necessary for their sterilization. I possess, for example, two test-tubes, containing the same infusion and associated with the same closed chamber, one of which has been heated five times and the other six. The former is quite turbid, while the latter is perfectly clear. In this case five heatings had left some of the more resistant germs still unkilled, which were destroyed by the sixth heating. Of two other tubes charged with a different infusion, one has been heated seven times and is now full of life; the other has been heated eight times and is perfectly barren.

With due care the method of sterilization here described is infallible, however highly infective the surrounding atmosphere may be. But here, as elsewhere in these difficult inquiries, the sagacity which comes in great part from nature, the skill which comes from training, and the care which ought to root itself in his moral constitution, are all necessary to save the experimenter from error and to lead him to the truth.

§ 23. *Mortality of Germs through defect of Oxygen produced by Exhaustion with the Sprengel Pump.*

An equally striking mode of sterilization is now to be described. The crowding together of the organisms so as to form in a multitude of cases a heavy, corrugated, fatty scum upon the surface of the infusions obviously indicated that air was a necessity of their life. In some cases the oxygen dissolved in the infusions sufficed to enable the *Bacteria* to cloud them from top to bottom; but in many cases they gathered at the top, and formed

there a living layer through which no oxygen could pass to the liquid underneath, which, thus surmounted, remained as clear as water. The observation of these facts, and many others of a similar bearing, suggested inquiry into the effect which the more or less perfect withdrawal of the air from the infusions would have upon the development of life.

A few experiments with an ordinary air-pump were, in the first instance, made. The necks of a series of pipette bulbs charged with turnip-infusion were drawn out at the middle to a tube of very small diameter. The open end of the neck being connected with the air-pump, the bulbs were exhausted. In some cases, to render the removal of the air more perfect, hydrogen was admitted into the bulb and was afterwards withdrawn by the air-pump. Before they were detached from the pump the bulbs were immersed in lukewarm water. They boiled freely, and after a minute's ebullition the narrowed necks were hermetically sealed. The bulbs were then submerged in cold water, which was gradually raised to 212° F. and kept boiling for ten minutes; they were afterwards removed and placed in a room with a temperature of about 90° Fahr.

Four bulbs were thus treated in a preliminary experiment on the 7th of March. Two of them remain crystal clear to the present hour; the two others became cloudy, but remained entirely free from scum. The cloudiness, I may add, was barely perceptible, but it was perfectly distinct to the practised eye.

By such means, however, the removal of the air must have been more or less imperfect, and I therefore resorted to the far more effective Sprengel pump. To connect them with the pump, the bulbs were thrown into the form represented in fig. 21. After the neck of the bulb had been plugged with cotton-wool it was bent

at right angles above the plug, and a portion of it was drawn out to a tube of capillary diameter, represented at *o*. The end *a* was connected with the Sprengel pump, and after the exhaustion had been continued for the required interval, the neck of the bulb was sealed at *o*.

On the 14th of March three bulbs charged with turnip-infusion, from which the air had been as far as possible removed by the ordinary air-pump, were connected with the Sprengel, which continued the exhaustion uninterruptedly for three hours. The air dissolved in the liquid freely escaped from it at first, and it continued to appear in minute bubbles long after the exhaustion had reached a considerable degree of perfection. The drawn-out necks of the bulbs being hermetically sealed, the infusion within them was maintained as before for ten minutes at the boiling temperature.

It will be remembered that when the infusion and the air above it possessed their ordinary supply of oxygen, 180 minutes' boiling failed to sterilize the turnip-infusion. Here, when the air was withdrawn from the liquid, exposure for one-eighteenth of the foregoing interval sufficed to produce perfect barrenness. The infusion in the three bulbs here operated on remains to the present hour clear in body and perfectly free from scum.

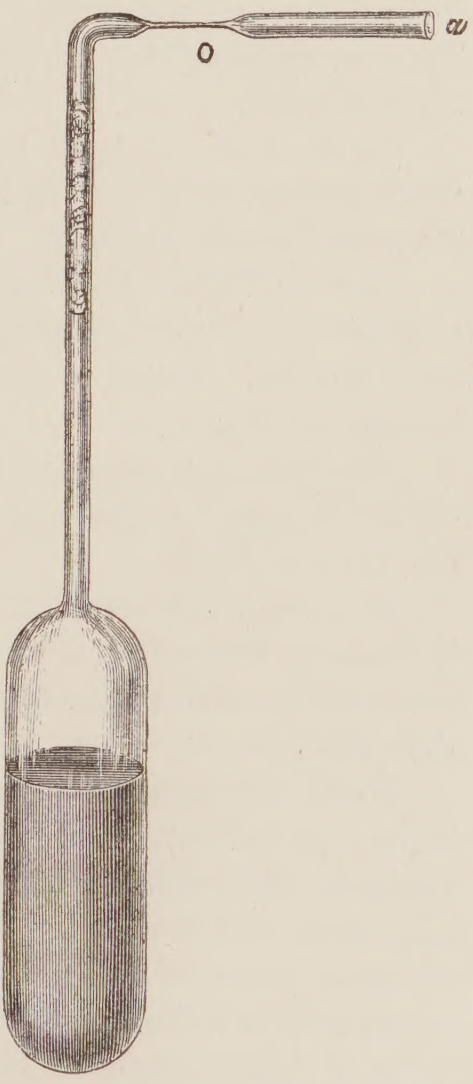
On the 15th of March seven bulbs charged with infusion of turnip were treated in the manner just described, being purged of their air by three hours' action of the Sprengel pump, and boiled for ten minutes afterwards. Six out of the seven remain perfectly pellucid.

On the 16th of March the result was still further verified. Seven bulbs were then charged with turnip-infusion, exhausted first by the air-pump, and afterwards by five hours' action of the Sprengel pump. Hermetic-

ally sealed and boiled as before, six out of the seven remain as clear as distilled water.

On the 20th of March seven bulbs were charged with infusion of cucumber, and subjected to the action of the Sprengel pump for seven hours. They were afterwards treated in the manner just described. They were all completely sterilized.

Fig. 21.



On the 27th of March three bulbs were charged with cucumber-infusion and subjected to the action of the Sprengel pump for five hours. One of them was subsequently boiled for five minutes, another for one minute, while the third was left unboiled. This third tube became faintly cloudy, but the two others remain perfectly free from life.

This result invited repetition. On the 29th accordingly six bulbs of cucumber-infusion were exhausted for five hours, and afterwards sealed and boiled for a single minute. Five of the six bulbs remained permanently clear; one became cloudy.

On the 30th of March six bulbs containing turnip-infusion were exhausted for five hours and boiled afterwards for a minute. Five remain perfectly clear, one has become muddy.

On the 6th of April five bulbs of beef-infusion were subjected for three hours to the action of the Sprengel pump and boiled for a minute afterwards. They all remain brilliant.

On the 7th of April five bulbs of mutton-infusion were treated like the beef-bulbs, being exhausted for three hours and boiled for a minute. All remain clear. This experiment was repeated and confirmed on the 20th of April.

On the 14th of April three bulbs of pork-infusion were exhausted for four hours and boiled for a minute. They all remain pellucid.

On the 17th of April four bulbs of accurately neutralized urine were exhausted for five hours and boiled for a minute. Three of them remain bright; one has become cloudy.

This does not exhaust the list of instances. Many other infusions have been sterilized by this method since the 17th of April.

It is perfectly certain that in most, if not all, of these cases 200 minutes' boiling would have proved insufficient to sterilize the infusion if it had been supplied with air.

Here the question naturally arises:—What would happen if the bulbs were exhausted and left unboiled? Probably with sufficiently perfect exhaustion all infusions would be sterilized. But in the trials I have thus far made some of the unboiled infusions have become cloudy, while others have remained clear. Thus three bulbs of mutton-infusion exhausted for four hours, two bulbs of beef-infusion exhausted for three hours, four bulbs of pork-infusion exhausted for four hours and left unboiled remain as transparent and as free from life as their boiled companions. Various other instances of sterilization without boiling might be cited. On the

other hand, three bulbs of neutralized urine, exhausted for five hours and left unboiled, became cloudy. A case of cucumber-infusion which behaved similarly has been cited above. It is difficult, if not impossible, to remove from the infusion and the space above it the last traces of air; and when backed by a highly nutritive liquid an infinitesimal residue of oxygen can develop a sensible amount of life. I may add that I have tested the exhaustion of some of the cloudy bulbs, and have found it in every case defective.

The foregoing instances sufficiently illustrate the dependence of the organisms here under review upon the supply of oxygen.[1] I think it probable that the principle thus indicated is capable of useful and extensive practical application.

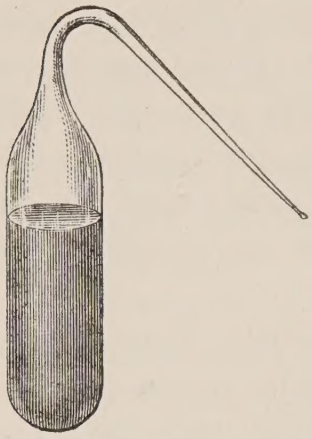
§ 24. *Mortality of Germs through defect of Oxygen consequent on boiling the Infusion.*

Long prior to these experiments with the Sprengel pump, the influence of atmospheric oxygen on the life of these organisms had been brought home to me. It revealed itself in a striking manner in experiments with infusions purged of air by boiling, the vessels containing them being carefully sealed during ebullition. At a time when the atmosphere of our laboratory was so laden with infection that no escape for animal or vegetable liquids introduced in the usual way into closed chambers was possible, it was perfectly easy to keep the same infusions pellucid for an indefinite time in vessels purged of air by boiling and properly sealed. I will give a few

[1] In search of this gas they sometimes rise into the liquid film which covers the interior of the bulb to the height of an inch and more above the surface of the liquid, forming within the bulb a gauzy scum which appears as if lifted by capillary attraction.

out of the multitude of examples that might be cited in proof of this statement.

Fig. 22.



On the 2nd of October fourteen of our ordinary retort-flasks (fig. 22) were charged with a neutralized infusion of hay. They were boiled for three minutes in an oil-bath, and hermetically sealed whilst boiling. Thirteen out of the fourteen tubes remained perfectly barren, retaining for months their pristine colour and transparency.

On the 18th of November six retort-flasks were filled with turnip-, five with cucumber-, five with beetroot-, and four with parsnep-infusion. The six turnip-flasks remained permanently pellucid, yielding a clear water-hammer ring. The five beetroot-flasks remained also permanently barren, all yielding the water-hammer sound. Of the parsnep-flasks, two became turbid, but two remained clear. Of the cucumber-flasks, three became cloudy, while three remained permanently clear. Neither in the case of the parsnep nor in that of the cucumber could the water-hammer sound be obtained from the cloudy flasks, and when their sealed ends were broken under water, the vacuum was found defective. In the clear tubes, on the contrary, it was found practically perfect.

Again, on the 20th of November, seventeen retort-flasks were charged with infusions of turnip, cucumber, and parsnep. They were boiled for three minutes in an oil-bath, and carefully sealed while boiling. The six turnip-flasks remained permanently clear, maintaining for months their sharp water-hammer sound. Of the five parsnep-flasks, one became turbid and four remained

permanently clear. These latter only yielded the water-hammer sound. The turbid flask, on the contrary, when shaken yielded no such sound, and when its sealed end was broken under water, its vacuum proved defective. Of the six cucumber-flasks, two became turbid, the remaining four being perfectly clear. On breaking their sealed ends under water, one-third of one of the turbid flasks and one-fourth of the other remained unfilled by the liquid.

On the 6th of December eighteen retort-flasks were charged with cucumber-infusion. They were boiled for the usual time, that is three minutes, extreme care being taken to seal them during the issue of the steam. The water-hammer sound in all these flasks was particularly sharp and clear. Exposed to a temperature of 90° Fahr. for many weeks, seventeen of them remained perfectly pellucid; while the same infusion in a sealed bulb with filtered air above it, and dissolved in it, swarmed with life after boiling for sixty times the interval here found effectual.

On the day subsequent to its preparation, one of these well-exhausted flasks was unexpectedly found turbid and covered with scum. But on examining the sealed end it was found snipped off. The laboratory air had thus entered the flask and given birth to the observed organisms. Failures of this sort have a demonstrative force greater even than successes; they render so obviously plain the external source of the contamination.

It is worth saying here that the observation just recorded was of frequent occurrence. The fineness of the sealed points of our retort-flasks renders them very liable to be snipped off if they are not handled with care. After preparation they are usually suspended on a wire or on a wooden support; and in frequent instances, after such suspension, I have found a flask

differentiating itself by thick turbidity from a number of perfectly pellucid neighbours, the yielding of the flask being immediately traced to the fracture of its sealed end.

With the view of showing how readily, unless extreme care is taken, contamination may enter hermetically-sealed vessels, the following experiment was made on the 6th of December. Four retort-flasks were charged with cucumber-infusion, boiled for the usual time and sealed, not during the outrush of the steam, but a moment after ebullition had ceased. On the 9th of December three of these four flasks were faintly but distinctly turbid. The reason is obvious. On the cessation of the ebullition, a momentary condensation of the steam above the infusion caused an indraught—slight, no doubt, but still sufficient to contaminate or vivify the infusion.

The source of the contagium was also indicated by the following experiments. A large number of retort-flasks, embracing infusions of snipe, wild duck, partridge, hare, rabbit, mutton, turbot, salmon, whiting, mullet, turnip, and hay, had remained over from my stock of 1875. After a year's exposure to the temperature of our warm room not one of these flasks showed the slightest trace of turbidity or life. On the 7th of December the sealed ends of forty of them were snipped off in the laboratory. Five days afterwards twenty-seven of them were found swarming with organisms—a considerably higher percentage than that obtained by the same process in the same laboratory a year previously.

It is needless to dwell with any emphasis on the obvious inference from all this, namely, that the contagium is external to the infusions, that it is something in the air, and that at different times we have different

amounts of aerial interspace free from the floating contagium.

§ 25. *Critical Review of the last two Sections.*

It has been my desire and aim throughout this inquiry to free it as much as possible from uncertainty and doubt. I have tried to render the facts safe by laborious repetition, and the interpretations of those facts secure by close and constant criticism. Thus, in reference to our present subject, I had to put to myself very definitely the question whether the permanent clearness of an infusion exposed to a very moderate amount of heat, after having been freed from air by boiling or by the Sprengel pump, was really due to the destruction of the germs in the infusion. Even in a highly infective atmosphere, from three to five minutes' boiling in an oil-bath suffices to sterilize our retort-flasks, while it is perfectly certain that exposure to the boiling temperature for fifty times this interval may fail to kill the germs of an infusion containing a good supply of atmospheric air. A similar remark applies to our experiments with the Sprengel pump. I asked myself whether in these cases the life of the germs was not suspended merely, instead of being destroyed. It was quite conceivable that germs endowed with vital power, ready to act under proper conditions, might still exist in our hermetically-sealed flasks, although the entire absence of oxygen rendered their further development impossible.

That something more than a mere temporary hindrance to development is here involved was, however, proved by many of the experiments just recorded. These experiments showed that after hermetically-sealed flasks had remained pellucid, not only for days but for

weeks and months, and in some cases for more than a year, on their sealed ends being broken off, even in ordinary air, they by no means invariably showed signs of life afterwards. Many of them remained permanently barren while copiously supplied with oxygen.

Special experiments were, however, made to illustrate this point. First of all, as I have already recounted, hermetically-sealed retort-flasks were opened in one of our lower store-rooms, and though supplied with oxygen from this source they showed subsequently no signs of life. A considerable number of retort-flasks had also their sealed ends broken off in the midst of a spirit-lamp flame. It is known that gunpowder can be dropped through such a flame without ignition; and in a few rare instances the infusions which had their first supply of air thus passed through a flame showed subsequent signs of life. In such cases the germinal matter had been sucked so rapidly through the flame that it escaped destruction; but in the vast majority of instances the sterilized infusions remained sterile.

Mechanical arrangements were also made for the breaking off of the sealed ends in a receiver filled with filtered air. But it is by no means easy to perfectly cleanse from infectious matter the instruments used in such experiments, though with sufficient practice this might certainly be done. The consequence was that some of the flasks opened in filtered air yielded subsequently. The following is an illustrative case:—On the 3rd of January ten flasks were charged with cucumber-, turnip-, artichoke-, and melon-infusions. They were boiled for the usual time, sealed during ebullition, and exposed afterwards to a warm temperature. Their sealed ends were then broken off by a mechanical contrivance placed in a receiver containing filtered air. The two artichoke-flasks remained permanently barren after-

wards. Of the two melon-flasks, one remained barren and the other developed life. Of the two cucumber-flasks, one became turbid, the other remained clear. Of the four turnip-flasks, two became turbid and two remained clear. Out of the ten flasks, therefore, all freely connected with the external air, six remained permanently barren. By further practice barrenness in almost every instance has been secured. The conclusion, I think, is obvious. It is not the heat alone that destroys these germs, for fifty times the amount of heat will not accomplish this when oxygen in due quantity is present: the heat must be aided by the withdrawal of the oxygen.

§ 26. *Mortality of Germs through excess of Oxygen.*

The foregoing remarks lead naturally to a brief reference to the important experiments of M. Paul Bert[1] on the toxic influence of compressed oxygen. From the imperfect account of these experiments which first reached me, I inferred that the germs of putrefaction had been destroyed by mere mechanical pressure, and more than a year and a half ago I placed turnip-infusions in strong iron bottles, and subjected them for several days to an air-pressure of twenty-three atmospheres. When taken from the bottles the infusions were found one and all swarming with life. Last October I made a series of similar experiments with infusions of hay and turnip, subjecting them for several days to an air-pressure of twenty-seven atmospheres. When taken from their iron bottles the infusions were found one and all teeming with *Bacteria*.

I then resorted to pure oxygen, and found the same to be true of my infusions that M. Paul Bert had found

[1] Comptes Rendus, vol. lxxx. p. 1579.

true of his flesh, moist bread, boiled starch, strawberries, cherries, wine, and urine. Pressures varying from twenty-seven atmospheres to ten atmospheres of oxygen were employed. In all cases, however long the pressure was continued, or however favourable to putrefaction the surrounding temperature might be, the infusions (which embraced those of beef, mutton, and turnip) were found, when taken from the bottles, as clear as crystal and entirely free from life. It required, indeed, long subsequent exposure to the common air to infect infusions which had been thus surcharged with oxygen. Other bottles containing the same infusions were simultaneously subjected to a like pressure, not of oxygen, however, but of atmospheric air. When removed from the bottles they were one and all found in a state of putrefaction and swarming with life.

Thus when oxygen is wholly withdrawn from organic infusions, the life with which we are here concerned ceases. When, on the other hand, the gas is in considerable excess, it becomes a deadly poison to organisms which, in moderate quantities, it sustains. As in the case of temperature, so in regard to the supply of oxygen, there is a medial zone favourable to the play of vitality, beyond which, on both sides, life cannot exist.

The present memoir virtually ends here; but I will append a few brief sections which, though incomplete, are not without instruction.

§ 27. *Experiments on neutralized Urine.*

I have already communicated to the Royal Society the result of some experiments made with this liquid,[1] in which the potash employed for neutralization was subjected to a temperature of 220° Fahr. The alkali

[1] Proceedings, vol. xxv. p. 457.

was contained in tubes drawn out finely at the end, which were introduced into the flasks containing the urine, and broken by shaking after the acid urine had been completely sterilized by heat. The cases were exceedingly rare in which life showed itself in the urine thus neutralized. The preponderance of sterilized flasks over unsterilized ones was very great.

In the experiments now to be glanced at, neither the urine nor the potash was raised to a temperature higher than 212°. Wishing to ascertain how the refractory germs of our laboratory would fare in neutralized urine, on the 16th of February I had five pipette-bulbs charged with the liquid. It was neutralized by caustic potash, which on boiling produced copious precipitation. It was afterwards filtered and rendered very transparent. The bulbs had been well cleansed, filled with one-third of an atmosphere of filtered air, hermetically sealed, and exposed afterwards to the heat of a Bunsen flame. They were charged with the urine by breaking off their finely drawn out points in the body of the liquid. They were then again sealed, and subjected to the boiling temperature for ten minutes

Not one of these bulbs remained sterile. Two days subsequent to their preparation they were all swarming with organisms.

Three other bulbs were on the same occasion charged with precisely the same infusion, only instead of being associated with air they were well purged of air by five minutes' boiling in an oil-bath. While the steam was issuing they were hermetically sealed.

Not one of these bulbs has proved fruitful. They are all as brilliant and as free from life as they were after they had passed through the filter.

The difference here indicated is worthy of notice. In the one case five minutes' action completely steril-

izes; in the other ten minutes' action fails to do so. This latter interval indeed might be multiplied twenty-fold and still prove ineffectual. In the one case the process of boiling purged the liquid of its air; in the other case the air was retained within and above the liquid. The case therefore connects itself with our former illustrations.

On the 21st of February six bulbs were charged with fresh urine carefully neutralized and boiled for five minutes in an oil-bath. Of the six flasks, four remain perfectly clear and brilliant, one is slightly cloudy, and one turbid.

The urine here referred to was neutralized in our own laboratory; but as the importance of accurate neutralization has been much insisted on, I wished to check myself. At my request, therefore, Dr. Debus was good enough to send me from Greenwich a quantity of urine carefully neutralized by him. On the 1st of March seven retort-flasks were charged with the neutralized liquid. These were boiled for five minutes in an oil-bath and sealed during ebullition. Three of these flasks have become turbid, but four remain perfectly clear.

On the 5th of March Dr. Williamson was good enough to send me a supply of neutralized urine collected in a public urinal in University College. The colour was very deep, the odour was very bad, and the precipitation on boiling very copious. Fourteen retort-flasks were charged with this liquid on the 6th of March. Seven of them have gone bad, but seven of them remain clear.

On the 10th of March Dr. Frankland was good enough to send me a supply of urine neutralized by himself. It was introduced into four retort-flasks, which, like the others, were boiled for five minutes in hot oil and sealed during ebullition. None of these flasks have

shown the slightest sign of yielding. The liquid within all of them is as brilliant as it was when first introduced.

In every case here mentioned the liquid, after boiling, was exposed for several days to a temperature of 50° C.

The conflict described in the foregoing pages and the search for principles to reconcile the results occupied me too long to permit of my doing more than break ground on the subject of urine. I entertain, however, a strong opinion that by a little practice with this liquid, its sterilization by five minutes' boiling might be rendered certain in every case. As the experiments stand, they sufficiently negative the conclusion that neutralized urine furnishes a specially convincing illustration of spontaneous generation.

§ 28. *Hermetically-sealed Flasks exposed to the Sun of the Alps.*

A remark of Dr. Bastian's, wherein he refers to the power of the actinic rays of the sun to promote spontaneous generation,[1] caused me to take with me to Switzerland a number of flasks hermetically sealed with special care and charged with infusions of various kinds. Eighty of them were carefully packed in sawdust in London; but on my arrival at the Bel Alp, which stands at an elevation of some 7,000 feet above the sea, I found only forty-five of them unbroken. They were thus distributed:—

Beef	12	flasks.
Mackerel	12	„
Turnip	12	„
Fowl	9	„

[1] 'Nature,' vol. iii. p. 247.

For ten days of the splendid summer with which we were favoured during a portion of last July, these flasks were exposed daily to the sunlight upon the roof of the Bel Alp hotel. The sky during many of these days was of a deep and cloudless blue; and certainly in London the actinic rays never approached the power of those here brought to bear upon the infusions. The temperature for many hours of each day was about 120° Fahr. Every evening, when the thermometer had fallen to about 70°, the flasks were removed and suspended above the kitchen-range of the hotel, the temperature generally varying throughout the night from 70° to 80° Fahr. Such variations of temperature, it may be remarked, are deemed favourable to spontaneous generation.

After the sunny weather had disappeared, the flasks were allowed to remain for three weeks suspended in the kitchen, with occasional exposures to the sun; the average temperature of the kitchen where the flasks were hung was about 90° F. The result of the observations was that not one of these forty-five flasks yielded the slightest evidence of spontaneous generation. From first to last they all continued as limpid as distilled water.

The sealed ends of these flasks were afterwards nipped off under various circumstances, some on the Sparrenhorn, some on the glacier, some in the Massa Gorge, some amid the hair of my own head, and some in the rooms of the hotel. Many of them, moreover, were infected with water of various kinds—spring-water, lake-water, and glacier-water. It is not my object to give a detailed account of these experiments, but simply to say that it was not lack of nutritive power on the part of the infusions which prevented the appearance of organisms in the first instance; for when brought into

contact with infectious matter every one of the flasks showed its power of sustaining and multiplying life.

§ 29. *Remarks on Hermetic Sealing.*

A few brief remarks on this subject may, I think, be fitly interpolated here. Hermetic sealing during ebullition is an operation requiring some apprenticeship to perform it aright. The neck of the flask ought to be so narrow that the pressure of the steam within shall be always sensibly greater than that of the atmosphere without. This condition would be readily fulfilled if the liberation of the steam were absolutely uniform, and not by fits and starts. But it never is uniform, and if the channel through which the steam issues be wide, it is scarcely possible to avoid regurgitation. Sometimes the pressure within is above that of the atmosphere, and steam freely issues; but at the next moment, through liquid adhesion to the flask, and partial condensation above, the internal pressure may be below that of the atmosphere and permit air to enter. This alternate triumph of the inner and the outer pressure may be rendered plainly evident by the motions of the water condensed in the neck of the flask. The liquid acts as an index which moves to and fro, sometimes forward, sometimes backward, as the pressure varies. It is quite evident that contamination may be, and it is quite certain that contamination has been, thus introduced into flasks reputed to be free from air.

Even with considerable care and fairly disciplined manipulatory skill success is not invariable. Ten per cent. is not at all a large allowance to set down as defective in ordinary hermetically-sealed flasks. The recent opening of about two hundred flasks employed in my earlier experiments, under water and under caustic-

potash solution, furnishes the basis of this conclusion. Even in a comparatively pure atmosphere success does not in every instance attend the experimenter. At Kew, for example, on the 8th of January, thirteen retort-flasks were charged with infusions of cucumber, melon, beef, and sole. Twelve out of the thirteen remained perfectly limpid, but one of them (a cucumber-flask) became distinctly cloudy, and this one alone refused, when tested, to yield the water-hammer sound. With due training, however, success may be rendered invariable.

§ 30. *Experiments with Turnip-cheese Infusions.*

I am unwilling to omit all reference to experiments which have cost considerable labour, and which, though they have not been repeated and controlled to the extent that I could wish, contribute nevertheless to our knowledge of the present question. This unwillingness causes me to introduce here, in the briefest manner possible, a reference to a series of experiments made with turnip-cheese infusions, which have been so frequently cited as offering a conspicuous proof of the doctrine of spontaneous generation.

The experiments to which I refer were made in part with closed chambers, and in part with hermetically-sealed retort-flasks. The specific gravity of the infusions varied from 1008 to 1012. The cheeses employed were Cheshire, Cheddar, Gloucester, Dutch cheese, American cheese, Roquefort, and Parmesan, the quantity varying from half a grain to two grains for every ounce of the infusion. The cheese being first well triturated in a mortar, so as to render its particles very minute, was intimately mixed with the infusion, which was then boiled for a few minutes and passed through a filter. The filtered liquid was then introduced into its closed chamber, and boiled there for five minutes.

Sixteen such chambers were employed, one of them containing twelve test-tubes, each of the others only three. There were therefore fifty-seven test-tubes in all. The result of the experiment was, that out of the fifty-seven tubes, twenty-seven became turbid in a few days, while thirty remained for months without sensible alteration.[1]

A considerable number of retort-flasks were charged at the same time with the same infusions, and boiled for five minutes in an oil-bath. The great majority of these flasks remained perfectly intact.

Here, then, as elsewhere, the ground on which the doctrine of spontaneous generation has sought to plant itself slips from under it; for assuredly the scientific mind will attribute to other causes than to it the production of organisms in the minority of cases above referred to.

One likely cause may here be signalized and illustrated. The experiments of Spallanzani on the action of heat upon seeds are well known, and they have been frequently cited in support of the thesis that the briefest exposure to the temperature of 212° Fahr. suffices to destroy all living matter. I have repeated many of Spallanzani's experiments, and will here briefly refer to one series which bear upon the present point. Peas, kidney-beans, cress- and mustard-seed were tied up in small calico bags, and boiled for intervals varying from half a minute to five minutes. They were then carefully sown in flower-pots filled with well-prepared earth, and placed in a shed kept at a

[1] These chambers were prepared and their tubes charged prior to the introduction of hay into our laboratory last autumn, otherwise the immunity of a single one of them could not have been secured. The chambers employed had stood over from my last investigation, and no pains had been taken to render them air-tight.

temperature of 70° Fahr. An unboiled sample of every seed was sown at the same time beside the boiled ones. The unboiled seeds sprouted vigorously. Thirty seconds' exposure to the boiling temperature deprived both the peas and the beans of their power of germination. A few of the cress-seeds exposed for this interval sprouted, but the majority were killed, and all were killed by a minute's boiling. On the other hand, a very large proportion of the mustard-seeds boiled for thirty seconds germinated. The time of exposure in the case of this seed was doubled, trebled, and quadrupled, leaving still a residue of life. The fertile mustard-seeds gradually diminished in number as the time of boiling increased, but even after two minutes' boiling many of them germinated.

And now comes a fact which I deem of some importance as regards the present inquiry. When the calico bag was abandoned, and the mustard-seeds were placed loosely in water, so as to ensure not only the free communication to them of its temperature, but free diffusion between the soluble portions of the seeds and the surrounding liquid, not one of them escaped the ordeal of thirty seconds' boiling. In the first series of experiments, the bag which held the seeds together not only exercised a protecting influence itself, but it enabled the outside seeds to act as shields to the inside ones. Assuredly in a far higher degree will cheese shield germs contained within it. Unlike fruit and meat, it is highly impervious to water. It thus wards off the liquid on which the softening and swelling of the germ depend, so that within such a substance the life of a germ might be indefinitely prolonged.

FERMENTATION, AND ITS BEARINGS ON SURGERY AND MEDICINE.[1]

IV.

One of the most remarkable characteristics of the age in which we live, is its desire and tendency to connect itself organically with preceding ages—to ascertain how the state of things that now is came to be what it is. And the more earnestly and profoundly this problem is studied, the more clearly comes into view the vast and varied debt which the world of to-day owes to that fore-world, in which man by skill, valour, and well-directed strength first replenished and subdued the earth. Our prehistoric fathers may have been savages, but they were clever and observant ones. They founded agriculture, by the discovery and development of seeds whose origin is now unknown. They tamed and harnessed their animal antagonists, and sent them down to us as ministers, instead of rivals in the fight for life. Later on, when the claims of luxury added themselves to those of necessity, we find the same spirit of invention at work. We have no historic account of the first brewer, but we glean from history that his art was practised, and its produce relished, more than two thousand years ago. Theophrastus, who was born nearly four hundred years before Christ,

[1] A Discourse delivered before the Glasgow Science Lectures Association, October 19, 1876.

described beer as *the wine of barley*. It is extremely difficult to preserve beer in a hot country, still, Egypt was the land in which it was first brewed, the desire of man to quench his thirst with this exhilarating beverage overcoming all the obstacles which a hot climate threw in the way of its manufacture.

Our remote ancestors had also learned by experience that wine maketh glad the heart of man. Noah, we are informed, planted a vineyard, drank of the wine, and experienced the consequences. But, though wine and beer possess so old a history, a very few years ago no man knew the secret of their formation. Indeed, it might be said that until the present year no thorough and scientific account was ever given of the agencies which come into play in the manufacture of beer, of the conditions necessary to its health, and of the maladies and vicissitudes to which it is subject. Hitherto the art and practice of the brewer have resembled those of the physician, both being founded on empirical observation. By this is meant the observation of facts, apart from the principles which explain them, and which give the mind an intelligent mastery over them. The brewer learnt from long experience the conditions, not the reasons, of success. But he had to contend, and has still to contend, against unexplained perplexities. Over and over again his care has been rendered nugatory; his beer has fallen into acidity or rottenness, and disastrous losses have been sustained, of which he has been unable to assign the cause. It is the hidden enemies against which the physician and the brewer have hitherto contended, that recent researches are dragging into the light of day, thus preparing the way for their final extermination.

Let us glance for a moment at the outward and

visible signs of fermentation. A few weeks ago I paid a visit to a private still in a Swiss chalet; and this is what I saw. In the peasant's bedroom was a cask with a very large bunghole carefully closed. The cask contained cherries which had lain in it for fourteen days. It was not entirely filled with the fruit, an air-space being left above the cherries when they were put in. I had the bung removed, and a small lamp dipped into this space. Its flame was instantly extinguished. The oxygen of the air had entirely disappeared, its place being taken by carbonic acid gas.[1] I tasted the cherries: they were very sour, though when put into the cask they were sweet. The cherries and the liquid associated with them were then placed in a copper boiler, to which a copper head was closely fitted. From the head proceeded a copper tube which passed straight through a vessel of cold water, and issued at the other side. Under the open end of the tube was placed a bottle to receive the spirit distilled. The flame of small wood-splinters being applied to the boiler, after a time vapour rose into the head, passed through the tube, was condensed by the cold of the water, and fell in a liquid fillet into the bottle. On being tasted, it proved to be that fiery and intoxicating spirit known in commerce as Kirsch or Kirschwasser.

The cherries, it should be remembered, were left to themselves, no ferment of any kind being added to them. In this respect what has been said of the cherry applies also to the grape. At the vintage the fruit of the vine is placed in proper vessels, and abandoned to its own action. It ferments, producing carbonic acid; its sweetness disappears, and at the end of a certain

[1] The gas which is exhaled from the lungs after the oxygen of the air has done its duty in purifying the blood; the same also which effervesces from soda water and champagne.

time the unintoxicating grape-juice is converted into intoxicating wine. Here, as in the case of the cherries, the fermentation is spontaneous—in what sense spontaneous will appear more clearly by-and-by.

It is needless for me to tell a Glasgow audience that the beer-brewer does not set to work in this way. In the first place the brewer deals not with the juice of fruits, but with the juice of barley. The barley having been steeped for a sufficient time in water, it is drained and subjected to a temperature sufficient to cause the moist grain to germinate; after which, it is completely dried upon a kiln. It then receives the name of *malt*. The malt is crisp to the teeth, and decidedly sweeter to the taste than the original barley. It is ground, mashed up in warm water, then boiled with hops until all the soluble portions have been extracted; the infusion thus produced being called the *wort*. This is drawn off, and cooled as rapidly as possible; then, instead of abandoning the infusion, as the wine-maker does, to its own action, the brewer mixes yeast with his wort, and places it in vessels each with only one aperture open to the air. Soon after the addition of the yeast, a brownish froth, which is really new yeast, issues from the aperture, and falls like a cataract into troughs prepared to receive it. This frothing and foaming of the wort is a proof that the fermentation is active.

Whence comes the yeast which issues so copiously from the fermenting tub? What is this yeast, and how did the brewer become possessed of it? Examine its quantity before and after fermentation. The brewer introduces, say 10 cwts. of yeast; he collects 40, or it may be 50, cwts. The yeast has, therefore, augmented from four to five fold during the fermentation. Shall we conclude that this additional yeast has been spontaneously generated by the wort? Are we not rather re-

minded of that seed which fell into good ground, and brought forth fruit, some thirty fold, some sixty fold, some an hundred fold? On examination, this notion of organic growth turns out to be more than a mere surmise. In the year 1680, when the microscope was still in its infancy, Leeuwenhoek turned the instrument upon yeast, and found it composed of minute globules suspended in a liquid. Thus knowledge rested until 1835, when Cagniard de la Tour in France, and Schwann in Germany, independently, but animated by a common thought, turned microscopes of improved definition and heightened powers upon yeast, and found it budding and sprouting before their eyes. The augmentation of the yeast alluded to above was thus proved to arise from the growth of a minute plant now called *Torula* (or *Saccharomyces*) *Cerevisiæ*. Spontaneous generation is therefore out of the question. The brewer deliberately sows the yeast-plant, which grows and multiplies in the wort as its proper soil. This discovery marks an epoch in the history of fermentation.

But where did the brewer find his yeast? The reply to this question is similar to that which must be given if it were asked where the brewer found his barley. He has received the seeds of both of them from preceding generations. Could we connect without solution of continuity the present with the past, we should probably be able to trace back the yeast employed by my friend Sir Fowell Buxton to-day to that employed by some Egyptian brewer two thousand years ago. But you may urge that there must have been a time when the first yeast-cell was generated. Granted—exactly as there was a time when the first barley-corn was generated. Let not the delusion lay hold of you that a living thing is easily generated because it is small. Both the yeast-plant and the barley-plant lose themselves in the dim

twilight of antiquity, and in this our day there is no more proof of the spontaneous generation of the one than there is of the spontaneous generation of the other.

I stated a moment ago that the fermentation of grape-juice was spontaneous; but I was careful to add, 'in what sense spontaneous will appear more clearly by-and-by.' Now this is the sense meant. The wine-maker does not, like the brewer and distiller, deliberately introduce either yeast, or any equivalent of yeast, into his vats; he does not consciously sow in them any plant, or the germ of any plant; indeed, he has been hitherto in ignorance whether plants or germs of any kind have had anything to do with his operations. Still, when the fermented grape-juice is examined, the living *Torula* concerned in alcoholic fermentation never fails to make its appearance. How is this? If no living germ has been introduced into the wine-vat, whence comes the life so invariably developed there?

You may be disposed to reply, with Turpin and others, that in virtue of its own inherent powers, the grape-juice when brought into contact with the vivifying atmospheric oxygen, runs spontaneously and of its own accord into these low forms of life. I have not the slightest objection to this explanation, provided proper evidence can be adduced in support of it. But the evidence adduced in its favour, as far as I am acquainted with it, snaps asunder under the strain of scientific criticism. It is, as far as I can see, the evidence of men who, however keen and clever as *observers*, are not rigidly trained *experimenters*. These alone are aware of the precautions necessary in investigations of this delicate kind. In reference, then, to the life of the wine-vat, what is the decision of experiment when carried out by competent men? Let a quantity

of the clear, filtered 'must' of the grape be so boiled as to destroy such germs as it may have contracted from the air or otherwise. In contact with germless air the uncontaminated must never ferments. All the materials for spontaneous generation are there, but so long as there is no seed sown, there is no life developed, and no sign of that fermentation which is the concomitant of life. Nor need you resort to a boiled liquid. The grape is sealed by its own skin against contamination from without. By an ingenious device Pasteur has extracted from the interior of the grape its pure juice, and proved that in contact with pure air it never acquires the power to ferment itself, nor to produce fermentation in other liquids.[1] It is not, therefore, in the interior of the grape that the origin of the life observed in the vat is to be sought.

What then is its true origin? This is Pasteur's answer, which his well-proved accuracy renders worthy of all confidence. At the time of the vintage microscopic particles are observed adherent, both to the outer surface of the grape and to the twigs which support the grape. Brush these particles into a capsule of pure water. It is rendered turbid by the dust. Examined by a microscope, some of these minute particles are seen to present the appearance of organized cells. Instead of receiving them in water, let them be brushed into the pure inert juice of the grape. Forty-eight hours after this is done, our familiar *Torula* is observed budding and sprouting, the growth of the plant being accompanied by all the other signs of active fermentation. What is the inference to be drawn from this

[1] The liquids of the healthy animal body are also sealed from external contamination. Pure blood, for example, drawn with due precautions from the veins, will never ferment or putrefy in contact with pure air.

experiment? Obviously that the particles adherent to the external surface of the grape include the germs of that life which, after they have been sown in the juice, appears in such profusion. Wine is sometimes objected to on the ground that fermentation is 'artificial;' but we notice here the responsibility of nature. The ferment of the grape clings like a parasite to the surface of the grape; and the art of the wine-maker from time immemorial has consisted in bringing—and it may be added, ignorantly bringing—two things thus closely associated by nature into actual contact with each other. For thousands of years, what has been done consciously by the brewer, has been done unconsciously by the wine-grower. The one has sown his leaven just as much as the other.

Nor is it necessary to impregnate the beer-wort with yeast to provoke fermentation. Abandoned to the contact of our common air, it sooner or later ferments; but the chances are that the produce of that fermentation, instead of being agreeable, would be disgusting to the taste. By a rare accident we might get the true alcoholic fermentation, but the odds against obtaining it would be enormous. Pure air acting upon a lifeless liquid will never provoke fermentation; but our ordinary air is the vehicle of numberless germs which act as ferments when they fall into appropriate infusions. Some of them produce acidity, some putrefaction. The germs of our yeast-plant are also in the air; but so sparingly distributed that an infusion like beer-wort, exposed to the air, is almost sure to be taken possession of by foreign organisms. In fact, the maladies of beer are wholly due to the admixture of these objectionable ferments, whose forms and modes of nutrition differ materially from those of the true leaven.

Working in an atmosphere charged with the germs of these organisms, you can understand how easy it is to fall into error in studying the action of any one of them. Indeed it is only the most accomplished experimenter, who, moreover, avails himself of every means of checking his conclusions, that can walk without tripping through this land of pitfalls. Such a man the French chemist Pasteur has hitherto proved himself to be. He has taught us how to separate the commingled ferments of our air, and to study their pure individual action. Guided by him, let us fix our attention more particularly upon the growth and action of the true yeast-plant under different conditions. Let it be sown in a fermentable liquid, which is supplied with plenty of pure air. The plant will flourish in the aërated infusion, and produce large quantities of carbonic acid gas—a compound, as you know, of carbon and oxygen. The oxygen thus consumed by the plant is the free oxygen of the air, which we suppose to be abundantly supplied to the liquid. The action is so far similar to the respiration of animals, which inspire oxygen and expire carbonic acid. If we examine the liquid even when the vigour of the plant has reached its maximum, we hardly find in it a trace of alcohol. The yeast has grown and flourished, but it has almost ceased to act as a ferment. And could every individual yeast-cell seize, without any impediment, free oxygen from the surrounding liquid, it is certain that it would cease to act as a ferment altogether.

What, then, are the conditions under which the yeast-plant must be placed so that it may display its characteristic quality? Reflection on the facts already referred to suggests a reply, and rigid experiment confirms the suggestion. Consider the Alpine cherries in their closed vessel. Consider the beer in its barrel,

with a single small aperture open to the air, through which it is observed not to imbibe oxygen, but to pour forth carbonic acid. Whence come the volumes of oxygen necessary to the production of this latter gas? The small quantity of atmospheric air dissolved in the wort and overlying it would be totally incompetent to supply the necessary oxygen. In no other way can the yeast-plant obtain the gas necessary for its respiration than by wrenching it from surrounding substances in which the oxygen exists, not free, but in a state of combination. It decomposes the sugar of the solution in which it grows, produces heat, breathes forth carbonic acid gas, and one of the liquid products of the decomposition is our familiar alcohol. The act of fermentation, then, is a result of the effort of the little plant to maintain its respiration by means of *combined* oxygen, when its supply of free oxygen is cut off. As defined by Pasteur, fermentation is *life without air*.

But here the knowledge of that thorough investigator comes to our aid to warn us against possible error. It is not, he says, all yeast-cells that can thus live without air and provoke fermentation. They must be young cells which have caught their vegetative vigour from contact with free oxygen. But once possessed of this vigour the yeast, he alleges, may be transplanted into a saccharine infusion absolutely purged of air, where it will continue to live at the expense of the oxygen, carbon, and other constituents of the infusion. Under these new conditions its life, *as a plant*, will be by no means so vigorous as when it had a supply of free oxygen, but its action *as a ferment* will be indefinitely greater.

Does the yeast-plant stand alone in its power of provoking alcoholic fermentation? It would be sin-

gular if amid the multitude of low vegetable forms no other could be found capable of acting in a similar way. And here again we have occasion to marvel at that sagacity of observation among the ancients to which we owe so vast a debt. Not only did they discover the alcoholic ferment of yeast, but they had to exercise a wise selection in picking it out from others, and giving it special prominence. Place an old boot in a moist place, or expose common paste or a pot of jam to the air; it soon becomes coated with a blue-green mould, which is nothing else than the fructification of a little plant called *Penicillium glaucum*. Do not imagine that the mould has sprung spontaneously from boot, or paste, or jam; its germs, which are abundant in the air, have been sown, and have germinated, in as legal and legitimate a way as thistle-seeds wafted by the wind to a proper soil. Let the minute spores of *Penicillium* be sown in a fermentable liquid, which has been previously so boiled as to kill all other spores or seeds which it may contain; let pure air have free access to the mixture; the *Penicillium* will grow rapidly, striking long filaments into the liquid, and fructifying at its surface. Test the infusion at various stages of the plant's growth, you will never find in it a trace of alcohol. But forcibly submerge the little plant, push it down deep into the liquid, where the quantity of free oxygen that can reach it is insufficient for its needs, it immediately begins to act as a ferment, supplying itself with oxygen by the decomposition of the sugar, and producing alcohol as one of the results of the decomposition. Many other low microscopic plants act in a similar manner. In aërated liquids they flourish without any production of alcohol, but cut off from free oxygen they act as ferments, producing alcohol exactly as the real alcoholic leaven produces it,

only less copiously. For the discovery and apprehension of these facts we are indebted to Pasteur.

In the cases hitherto considered, the fermentation is proved to be the invariable correlative of *life*, being produced by organisms foreign to the fermentable substance. But the substance itself may also have within it, to some extent, the motive power of fermentation. The yeast-plant, as we have learned, is an assemblage of living cells; but so at bottom, as shown by Schleiden and Schwann, are all living organisms. Cherries, apples, peaches, pears, plums, and grapes, for example, are composed of cells, each of which is a living unit. And here I have to direct your attention to a point of extreme interest. In 1821, the celebrated French chemist, Bérard, established the important fact that all ripening fruits, exposed to the free atmosphere, absorb the oxygen of the atmosphere and liberate an approximately equal volume of carbonic acid. He also found that when ripe fruits were placed in a confined atmosphere, the oxygen of the atmosphere was first absorbed, and an equal volume of carbonic acid given out. But the process did not end here. After the oxygen had vanished, carbonic acid, in considerable quantities, continued to be exhaled by the fruits, which at the same time lost a portion of their sugar, becoming more acid to the taste, though the absolute quantity of acid was not augmented. This was an observation of capital importance, and Bérard had the sagacity to remark that the process might be regarded as a kind of fermentation.

Thus the living cells of fruits can absorb oxygen and breathe out carbonic acid, exactly like the living cells of the leaven of beer. Supposing the access of oxygen suddenly cut off, will the living fruit-cells as suddenly die, or will they continue to live as yeast lives, by

extracting oxygen from the saccharine juices round them? This is a question of extreme theoretic significance. It was first answered affirmatively by the able and conclusive experiments of Lechartier and Bellamy, and the answer was subsequently confirmed and explained by the experiments and the reasoning of Pasteur. Bérard only showed the absorption of oxygen, and the production of carbonic acid; Lechartier and Bellamy proved the production of alcohol, thus completing the evidence that it was a case of real fermentation, though the common alcoholic ferment was absent. So full was Pasteur of the idea that the cells of a fruit would continue to live at the expense of the sugar of the fruit, that once in his laboratory, while conversing on these subjects with M. Dumas, he exclaimed, 'I will wager that if a grape be plunged into an atmosphere of carbonic acid, it will produce alcohol and carbonic acid by the continued life of its own cells—that they will act for a time like the cells of the true alcoholic leaven.' He made the experiment, and found the result to be what he had foreseen. He then extended the inquiry. Placing under a bell-jar twenty-four plums, he filled the jar with carbonic acid gas; beside it he placed twenty-four similar plums uncovered. At the end of eight days, he removed the plums from the jar, and compared them with the others. The difference was extraordinary. The uncovered fruits had become soft, watery, and very sweet; the others were firm and hard, their fleshy portions being not at all watery. They had, moreover, lost a considerable quantity of their sugar. They were afterwards bruised, and the juice was distilled. It yielded six and a half grammes of alcohol, or one per cent. of the total weight of the plums. Neither in these plums, nor in the grapes first experimented on by Pasteur, could any trace of the

ordinary alcoholic leaven be found. As previously proved by Lechartier and Bellamy, the fermentation was the work of the living cells of the fruit itself, after air had been denied to them. When, moreover, the cells were destroyed by bruising, no fermentation ensued. The fermentation was the correlative of a vital act, and it ceased when life was extinguished.

Lüdersdorf was the first to show by this method that yeast acted, not, as Liebig had assumed, in virtue of its *organic*, but in virtue of its *organized* character. He destroyed the cells of yeast by rubbing them on a ground glass plate, and found that with the destruction of the organism, though its chemical constituents remained, the power to act as a ferment totally disappeared.

One word more in reference to Liebig may find a place here. To the philosophic chemist thoughtfully pondering these phenomena, familiar with the conception of molecular motion, and the changes produced by the interactions of purely chemical forces, nothing could be more natural than to see in the process of fermentation a simple illustration of molecular instability, the ferment propagating to surrounding molecular groups the overthrow of its own tottering combinations. Broadly considered, indeed, there is a certain amount of truth in this theory; but Liebig, who propounded it, missed the very kernel of the phenomena when he overlooked, or contemned, the part played in fermentation by microscopic life. He looked at the matter too little with the eye of the body, and too much with the spiritual eye. He practically neglected the microscope, and was unmoved by the knowledge which its revelations would have poured in upon his mind. His hypothesis, as I have said, was natural—nay, it was a striking illustration of Liebig's power to penetrate and unveil

molecular actions; but it was an error, and as such has proved an *ignis fatuus* instead of a *pharos* to some of his followers.

I have said that our air is full of the germs of ferments differing from the alcoholic leaven, and sometimes seriously interfering with the latter. They are the weeds of this microscopic garden which often overshadow and choke the flowers. Let us take an illustrative case. Expose milk to the air. It will, after a time, turn putrid or sour, separating like blood into clot and serum. Place a drop of such milk under a powerful microscope and watch it closely. You see the minute butter-globules animated by that curious quivering motion called the Brownian motion. But let not this attract your attention too much, for it is another motion that we have now to seek. Here and there you observe a greater disturbance than ordinary among the globules; keep your eye upon the place of tumult, and you will probably see emerging from it a long eel-like organism, tossing the globules aside and wriggling more or less rapidly across the field of the microscope. Part of the change wrought in the milk is due to this organism, which from its motions receives the name of *vibrio*. In curdled milk you find other organisms, small, motionless, and usually linked together like beads on a string. It is these which cause the milk to separate into clot and serum. They constitute the lactic ferment of milk, as the yeast-plant does the alcoholic ferment of sugar. But milk may become putrid without becoming sour. Examine putrid milk microscopically, and you find it swarming with shorter organisms, sometimes associated with the vibrios, sometimes alone, and often manifesting a wonderful alacrity of motion. Keep these organisms and their germs out of your milk and it

will never putrefy. Expose a mutton-chop to the air and keep it moist; in summer weather it soon stinks. Place a drop of the juice of the fetid chop under a powerful microscope; it is seen swarming with organisms resembling those in the putrid milk. These organisms, which receive the common name of *bacteria*,[1] are the agents of all putrefaction. Keep them and their germs from your meat and it will remain for ever sweet. Thus we begin to see that within the world of life to which we ourselves belong, there is another living world requiring the microscope for its discernment, but which, nevertheless, has the most important bearing on the welfare of the higher life-world.

And now let us reason together as regards the origin of these bacteria. A granular powder is placed in your hands, and you are asked to state what it is. You examine it, and have, or have not, reason to suspect that seeds of some kind are mixed up in it. To determine this point you prepare a bed in your garden, sow in it the powder, and soon after find a mixed crop of docks and thistles sprouting from your bed. Until this powder was sown neither docks nor thistles ever made their appearance in your garden. You repeat the experiment once, twice, ten times, fifty times. From fifty different beds after the sowing of the powder, you obtain the same crop. What will be your response to the question proposed to you? 'I am not in a condition,' you would say, 'to affirm that every grain of the powder is a dock-seed, or a thistle-seed; but I am in a condition to affirm that both dock and thistle-seeds form, at all events, part of the powder.' Supposing a succession of such powders to be placed in your hands with grains becoming gradually smaller, until they dwindle to the

[1] Doubtless organisms exhibiting grave specific differences are grouped together under this common name.

size of impalpable dust particles; assuming that you treat them all in the same way, and that from every one of them in a few days you obtain a definite crop—it may be clover, it may be mustard, it may be mignonette, it may be a plant more minute than any of these—the smallness of the particles, or of the plants that spring from them, does not affect the validity of the conclusion. Without a shadow of misgiving you would conclude that the powder must have contained the seeds or germs of the life observed. There is not in the range of physical science, an experiment more conclusive nor an inference safer than this one.

Supposing the powder to be light enough to float in the air, and that you are enabled to see it there just as plainly as you saw the heavier powder in the palm of your hand. If the dust sown by the air instead of by the hand produce a definite living crop, with the same logical rigour you would conclude that the germs of this crop must be mixed with the dust. To take an illustration: the spores of the little plant *Penicillium glaucum*, to which I have already referred, are light enough to float in the air. A cut apple, a pear, a tomato, a slice of vegetable marrow, or, as already mentioned, an old moist boot, a dish of paste, or a pot of jam, constitutes a proper soil for the *Penicillium*. Now, if it could be proved that the dust of the air when sown in this soil produces this plant, while, wanting the dust, neither the air, nor the soil, nor both together can produce it, it would be obviously just as certain, in this case, that the floating dust contains the germs of *Penicillium*, as that the powders sown in your garden contained the germs of the plants which sprung from them.

But how is the floating dust to be rendered visible? In this way. Build a little chamber and provide it

with a door, windows, and window-shutters. Let an aperture be made in one of the shutters through which a sunbeam can pass. Close the door and windows so that no light shall enter save through the hole in the shutter. The track of the sunbeam is at first perfectly plain and vivid in the air of the room. If all disturbance of the air of the chamber be avoided, the luminous track will become fainter and fainter, until at last it disappears absolutely, and no trace of the beam is to be seen. What rendered the beam visible at first? The floating dust of the air, which, when thus illuminated and observed, is as palpable to sense as dust or powder placed on the palm of the hand. In the still air the dust gradually sinks to the floor or sticks to the walls and ceiling, until finally, by this self-cleansing process, the air is entirely freed from mechanically suspended matter.

Thus, far, I think, we have made our footing sure. Let us proceed. Chop up a beefsteak and allow it to remain for two or three hours just covered with warm water; you thus extract the juice of the beef in a concentrated form. By properly boiling the liquid and filtering it, you can obtain from it a perfectly transparent beef-tea. Expose a number of vessels containing this tea to the moteless air of your chamber; and expose a number of vessels containing precisely the same liquid to the dust-laden air. In three days every one of the latter stinks, and examined with the microscope every one of them is found swarming with the bacteria of putrefaction. After three months, or three years, the beef-tea within the chamber, if properly sterilized in the first instance, will be found as sweet and clear, and as free from bacteria, as it was at the moment when it was first put in. There is absolutely no difference between the air within and that without, save that the one is dustless and the other dust-

laden. Clinch the experiment thus: Open the door of your chamber and allow the dust to enter it. In three days afterwards you have every vessel within the chamber swarming with bacteria, and in a state of active putrefaction. Here, also, the inference is quite as certain as in the case of the powder sown in your garden. Multiply your proofs by building fifty chambers instead of one, and by employing every imaginable infusion of wild animals and tame; of flesh, fish, fowl, and viscera; of vegetables of the most various kinds. If in all these cases you find the dust infallibly producing its crop of bacteria, while neither the dustless air nor the nutritive infusion, nor both together, are ever able to produce this crop, your conclusion is simply irresistible that the dust of the air contains the germs of the crop which has appeared in your infusions. I repeat there is no inference of experimental science more certain than this one. In the presence of such facts, to use the words of a paper lately published in the 'Philosophical Transactions,' it would be simply monstrous to affirm that these swarming crops of bacteria are spontaneously generated.

Is there then no experimental proof of spontaneous generation? I answer without hesitation, *none!* But to doubt the experimental proof of a fact, and to deny its possibility, are two different things, though some writers confuse matters by making them synonymous. In fact, this doctrine of spontaneous generation, in one form or another, falls in with the theoretic beliefs of some of the foremost workers of this age; but it is exactly these men who have the penetration to see, and the honesty to expose, the weakness of the evidence adduced in its support.

And here observe how these discoveries tally with

the common practices of life. Heat kills the bacteria, colds numbs them. When my housekeeper has pheasants in charge which she wishes to keep sweet, but which threaten to give way, she partially cooks the birds, kills the infant bacteria, and thus postpones the evil day. By boiling her milk she also extends its period of sweetness. Some weeks ago in the Alps I made a few experiments on the influence of cold upon ants. Though the sun was strong, patches of snow still maintained themselves on the mountain slopes. The ants were found in the warm grass and on the warm rocks adjacent. Transferred to the snow the rapidity of their paralysis was surprising. In a few seconds a vigorous ant, after a few languid struggles, would wholly lose its power of locomotion, and lie practically dead upon the snow. Transferred to the warm rock, it would revive, to be again smitten with death-like numbness when retransferred to the snow. What is true of the ant is specially true of our bacteria. Their active life is suspended by cold, and with it their power of producing or continuing putrefaction. This is the whole philosophy of the preservation of meat by cold. The fishmonger, for example, when he surrounds his very assailable wares by lumps of ice, stays the process of putrefaction by reducing to numbness and inaction the organisms which produce it, and in the absence of which his fish would remain sweet and sound. It is the astonishing activity into which these bacteria are pushed by warmth that renders a single summer's day sometimes so disastrous to the great butchers of London and Glasgow. The bodies of guides lost in the crevasses of Alpine glaciers have come to the surface forty years after their interment, without the flesh showing any sign of putrefaction. But the most astonishing case of this kind is that of the hairy elephant of Siberia which was

found incased in ice. It had been buried for ages, but when laid bare its flesh was sweet, and for some time afforded copious nutriment to the wild beasts which fed upon it.

Beer is assailable by all the organisms here referred to, some of which produce acetic, some lactic, and some butyric acid, while yeast is open to attack from the bacteria of putrefaction. In relation to the particular beverage the brewer wishes to produce, these foreign ferments have been properly called *ferments of disease.* The cells of the true leaven are globules, usually somewhat elongated. The other organisms are more less rod-like or eel-like in shape, some of them being beaded so as to resemble necklaces. Each of these organisms produces a fermentation and a flavour peculiar to itself. Keep them out of your beer and it remains for ever unaltered. Never without them will your beer contract disease. But their germs are in the air, in the vessels employed in the brewery; even in the yeast used to impregnate the wort. Consciously or unconsciously, the art of the brewer is directed against them. His aim is to paralyze, if he cannot destroy them.

For beer, moreover, the question of temperature is one of supreme importance; indeed, the recognized influence of temperature is causing on the Continent of Europe a complete revolution in the manufacture of beer. When I was a student in Berlin, in 1851, there were certain places specially devoted to the sale of Bavarian beer, which was then making its way into public favour. This beer is prepared by what is called the process of *low fermentation;* the name being given partly because the yeast of the beer, instead of rising to the top and issuing through the bunghole, falls to the bottom of the cask; but partly, also, because it is produced at a low temperature. The other and older

process, called *high fermentation*, is far more handy, expeditious, and cheap. In high fermentation eight days suffice for the production of beer; in low fermentation, ten, fifteen, even twenty days are found necessary. Vast quantities of ice, moreover, are consumed in the process of low fermentation. In the single brewery of Dreher, of Vienna, a hundred million pounds of ice are consumed annually in cooling the wort and beer. Notwithstanding these obvious and weighty drawbacks, the low fermentation is rapidly displacing the high upon the Continent. Here are some statistics which show the number of breweries of both kinds existing in Bohemia in 1860, 1865, and 1870:—

	1860.	1865.	1870.
High Fermentation . .	281	81	18
Low Fermentation . .	135	459	831

Thus in ten years the number of high-fermentation breweries fell from 281 to 18, while the number of low-fermentation breweries rose from 135 to 831. The sole reason for this vast change—a change which involves a great expenditure of time, labour, and money—is the additional command which it gives the brewer over the fortuitous ferments of disease. These ferments, which, it is to be remembered, are living organisms, have their activity suspended by temperatures below 10° C., and as long as they are reduced to torpor the beer remains untainted either by acidity or putrefaction. The beer of low fermentation is brewed in winter, and kept in cool cellars; the brewer being thus enabled to dispose of it at his leisure, instead of forcing its consumption to avoid the loss involved in its alteration if kept too long. Hops, it may be remarked, act to some extent as an antiseptic to beer. The essential oil of the hop is bactericidal; hence the strong impregnation with hop juice of all beer intended for exportation.

These low organisms, which one might be disposed to regard as the beginnings of life, were we not warned that the microscope, precious and perfect as it is, has no power to show us the real beginnings of life, are by no means purely useless or purely mischievous in the economy of nature. They are only noxious when out of their proper place. They exercise a useful and valuable function as the burners and consumers of dead matter, animal and vegetable, reducing such matter, with a rapidity otherwise unattainable, to innocent carbonic acid and water. Furthermore, they are not all alike, and it is only restricted classes of them that are really dangerous to man. One difference in their habits is worthy of special reference here. Air, or rather the oxygen of the air, which is absolutely necessary to the support of the bacteria of putrefaction, is, according to Pasteur, absolutely deadly to the vibrios which provoke the butyric acid fermentation. This has been illustrated by the following beautiful observation.

A drop of the liquid containing those small organisms is placed upon glass, and on the top is placed a circle of exceedingly thin glass; for, to magnify them sufficiently, it is necessary that the object-glass of the microscope should come very close to the organisms. Round the edge of the circular plate of glass the liquid is in contact with the air, and incessantly absorbs it, including the oxygen. Here, if the drop be charged with bacteria, we have a zone of very lively ones. But through this living zone, greedy of oxygen and appropriating it, the vivifying gas cannot penetrate to the centre of the film. In the middle, therefore, the bacteria die, while their peripheral colleagues continue active. If a bubble of air chance to be enclosed in the film, round it the bacteria will pirouette and wabble

until its oxygen has been absorbed, after which all their motions cease. Precisely the reverse of all this occurs with the vibrios of butyric acid. In their case it is the peripheral organisms that are first killed, the central ones remaining vigorous while ringed by a zone of dead. Pasteur, moreover, filled two vessels with a liquid containing these vibrios; through one vessel he led air, and killed its vibrios in half an hour; through the other he led carbonic acid, and after three hours found the vibrios fully active. It was while observing these differences of deportment fifteen years ago that the thought of life without air, and its bearing upon the theory of fermentation, flashed upon the mind of this admirable investigator.

We now approach an aspect of this question which concerns us still more closely, and will be best illustrated by an actual fact. A few years ago I was bathing in an Alpine stream, and returning to my clothes from the cascade which had been my shower-bath, I slipped upon a block of granite, the sharp crystals of which stamped themselves into my naked shin. The wound was an awkward one, but being in vigorous health at the time, I hoped for a speedy recovery. Dipping a clean pocket-handkerchief into the stream, I wrapped it round the wound, limped home, and remained for four or five days quietly in bed. There was no pain, and at the end of this time I thought myself quite fit to quit my room. The wound, when uncovered, was found perfectly clean, uninflamed, and entirely free from pus. Placing over it a bit of goldbeater's-skin, I walked about all day. Towards evening itching and heat were felt; a large accumulation of matter followed, and I was forced to go to bed again. The water-bandage was restored, but it was powerless to check

the action now set up; arnica was applied, but it made matters worse. The inflammation increased alarmingly, until finally I had to be carried on men's shoulders down the mountain and transported to Geneva, where, thanks to the kindness of friends, I was immediately placed in the best medical hands. On the morning after my arrival in Geneva, Dr. Gautier discovered an abscess in my instep, at a distance of five inches from the wound. The two were connected by a channel, or *sinus*, as it is technically called, through which he was able to empty the abscess, without the application of the lance.

By what agency was that channel formed—what was it that thus tore asunder the sound tissue of my instep, and kept me for six weeks a prisoner in bed? In the very room where the water dressing had been removed from my wound, and the goldbeater's-skin applied to it, I opened this year a number of tubes, containing perfectly clear and sweet infusions of fish, flesh, and vegetable. These hermetically-sealed infusions had been exposed for weeks, both to the sun of the Alps and to the warmth of a kitchen, without showing the slightest turbidity or sign of life. But two days after they were opened the greater number of them swarmed with the bacteria of putrefaction, the germs of which had been contracted from the dust-laden air of the room. And had the matter from my abscess been examined, my memory of its appearance leads me to infer that it would have been found equally swarming with these bacteria—that it was their germs which got into my incautiously opened wound, and that they were the subtile workers that burrowed down my shin, dug the abscess in my instep, and produced effects which might easily have proved fatal.

This apparent digression brings us face to face with

the labours of a man who combines the penetration of the true theorist with the skill and conscientiousness of the true experimenter, and whose practice is one continued demonstration of the theory that the putrefaction of wounds is to be averted by the destruction of the germs of bacteria. Not only from his own reports of his cases, but from the reports of eminent men who have visited his hospital, and from the opinions expressed to me by continental surgeons, do I gather that one of the greatest steps ever made in the art of surgery was the introduction of the antiseptic system of treatment, for which we are indebted to Professor Lister.

The interest of this subject does not slacken as we proceed. We began with the cherry-cask and beer-vat; we end with the body of man. There are persons born with the power of interpreting natural facts, as there are others smitten with everlasting incompetence in regard to such interpretation. To the former class in an eminent degree belonged the illustrious philosopher Robert Boyle, whose words in relation to this subject have in them the forecast of prophecy. 'And let me add,' writes Boyle in his 'Essay on the Pathological Part of Physik,' 'that he that thoroughly understands the nature of ferments and fermentations shall probably be much better able than he that ignores them, to give a fair account of divers phenomena of several diseases (as well fevers as others), which will perhaps be never properly understood without an insight into the doctrine of fermentations.'

Two hundred years have passed since these pregnant words were written, and it is only in this our day that men are beginning to fully realize their truth. In the domain of surgery the justice of Boyle's surmise has been most strictly demonstrated. But we now pass

the bounds of surgery proper, and enter the domain of epidemic disease, including those fevers so sagaciously referred to by Boyle. The most striking analogy between a *contagium* and a ferment is to be found in the power of indefinite self-multiplication possessed and exercised by both. You know the exquisitely truthful figures regarding leaven employed in the New Testament. A particle hid in three measures of meal leavens it all. A little leaven leaveneth the whole lump. In a similar manner, a particle of *contagium* spreads through the human body and may be so multiplied as to strike down whole populations. Consider the effect produced upon the system by a microscopic quantity of the virus of smallpox. That virus is, to all intents and purposes, a seed. It is sown as yeast is sown, it grows and multiplies as yeast grows and multiplies, and it always reproduces itself. To Pasteur we are indebted for a series of masterly researches, wherein he exposes the looseness and general baselessness of prevalent notions regarding the transmutation of one ferment into another. He guards himself against saying it is impossible. The true investigator is sparing in the use of this word, though the use of it is unsparingly ascribed to him; but, as a matter of fact, Pasteur has never been able to effect the alleged transmutation, while he has been always able to point out the open doorways through which the affirmers of such transmutations had allowed error to march in upon them.[1]

The great source of error here has been already alluded to in this discourse. The observers worked in

[1] Those who wish for an illustration of the care necessary in these researches, and of the carelessness with which they have in some cases been conducted, will do well to consult the Rev. W. H. Dallinger's excellent 'Notes on Heterogenesis' in the October number of the *Popular Science Review*.

an atmosphere charged with the germs of different organisms; the mere accident of first possession rendering now one organism, now another, triumphant. In different stages, moreover, of its fermentative or putrefactive changes, the same infusion may so alter as to be successively taken possession of by different organisms. Such cases have been adduced to show that the earlier organisms must have been transformed into the later ones, whereas they are simply cases in which different germs, because of changes in the infusion, render themselves valid at different times.

By teaching us how to cultivate each ferment in its purity—in other words, by teaching us how to rear the individual organism apart from all others,—Pasteur has enabled us to avoid all these errors. And where this isolation of a particular organism has been duly effected it grows and multiplies indefinitely, but no change of it into another organism is ever observed. In Pasteur's researches the Bacterium remained a Bacterium, the Vibrio a Vibrio, the Penicillium a Penicillium, and the Torula a Torula. Sow any of these in a state of purity in an appropriate liquid; you get it, and it alone, in the subsequent crop. In like manner, sow smallpox in the human body, your crop is smallpox. Sow there scarlatina, and your crop is scarlatina. Sow typhoid virus, your crop is typhoid—cholera, your crop is cholera. The disease bears as constant a relation to its contagium as the microscopic organisms just enumerated do to their germs, or indeed as a thistle does to its seed. No wonder then, with analogies so obvious and so striking, that the conviction is spreading and growing daily in strength, that reproductive parasitic life is at the root of epidemic disease—that living ferments finding lodgment in the body increase there and multiply, directly ruining the tissue on which they subsist, or

destroying life indirectly by the generation of poisonous compounds within the body. This conclusion, which comes to us with a presumption almost amounting to demonstration, is clinched by the fact that virulently infective diseases have been discovered with which living organisms are as closely and as indissolubly associated as the growth of Torula is with the fermentation of beer.

And here, if you will permit me, I would utter a word of warning to well-meaning people. We have now reached a phase of this question when it is of the very last importance that light should once for all be thrown upon the manner in which contagious and infectious diseases take root and spread. To this end the action of various ferments upon the organs and tissues of the living body must be studied; the habitat of each special organism concerned in the production of each specific disease must be determined, and the mode by which its germs are spread abroad as sources of further infection. It is only by such rigidly accurate inquiries that we can obtain final and complete mastery over these destroyers. Hence, while abhorring cruelty of all kinds, while shrinking sympathetically from all animal suffering—suffering which my own pursuits never call upon me to inflict,—an unbiassed survey of the field of research now opening out before the physiologist causes me to conclude, that no greater calamity could befall the human race than the stoppage of experimental inquiry in this direction. A lady whose philanthropy has rendered her illustrious said to me some time ago, that science was becoming immoral: that the researches of the past, unlike those of the present, were carried on without cruelty. I replied to her that the science of Kepler and Newton, to which she referred as moral, dealt with the laws and phenomena of inorganic nature; but that

one great advance made by modern science was in the direction of biology, or the science of life; and that in this new direction scientific inquiry, though at the outset pursued at the cost of some temporary suffering, would in the end prove a thousand times more beneficent than it had ever hitherto been. I said this because I saw that the very researches which the lady deprecated were leading us to such a knowledge of epidemic diseases as will enable us finally to sweep these scourges of the human race from the face of the earth.

This is a point of such capital importance that I should like to bring it home to your intelligence by a single trustworthy illustration. In 1850, two distinguished French observers, MM. Davainne and Rayer, noticed in the blood of animals which had died of the virulent disease called *splenic fever*, small microscopic organisms resembling transparent rods; but neither of them at that time attached any significance to the observation. In 1861, Pasteur published a memoir on the fermentation of butyric acid, wherein he described the organism which provoked it; and after reading this memoir it occurred to Davainne that splenic fever might be a case of fermentation set up within the animal body, by the organisms which had been observed by him and Rayer. This idea has been placed beyond all doubt by subsequent research.

Observations of the highest importance have also been made on splenic fever by Pollender and Brauell. Two years ago, Dr. Burdon Sanderson gave us a very clear account of what was known up to that time of this disorder. With regard to the permanence of the contagium, it had been proved to hang for years about localities where it had once prevailed; and this seemed to show that the rod-like organisms could not constitute the contagium, because their infective power

was found to vanish in a few weeks. But other facts established an intimate connexion between the organisms and the disease; so that a review of all the facts caused Dr. Sanderson to conclude that the contagium existed in two distinct forms: the one 'fugitive' and visible as transparent rods; the other permanent but 'latent,' and not yet brought within the grasp of the microscope.

At the time that Dr. Sanderson was writing this report, a young German physician, named Koch,[1] occupied with the duties of his profession in an obscure country district, was already at work, applying, during his spare time, various original and ingenious devices to the investigation of splenic fever. He studied the habits of the rod-like organisms, and found the aqueous humour of an ox's eye to be particularly suitable for their nutrition. With a drop of the aqueous humour he mixed the tiniest speck of a liquid containing the rods, placed the drop under his microscope, warmed it suitably, and observed the subsequent action. During the first two hours hardly any change was noticeable; but at the end of this time the rods began to lengthen, and the action was so rapid that at the end of three or four hours they attained from ten to twenty times their original length. At the end of a few additional hours they had formed filaments in many cases a hundred times the length of the original rods. The same filament, in fact, was frequently observed to stretch through several fields of the microscope. Sometimes they lay in straight lines parallel to each other, in other cases they were bent, twisted, and coiled into the most graceful figures; while sometimes they formed knots of such bewildering complexity that it was im-

[1] This, I believe, was the first reference to the researches of Koch made in this country. 1879.

possible for the eye to trace the individual filaments through the confusion.

Had the observation ended here an interesting scientific fact would have been added to our previous store, but the addition would have been of little practical value. Koch, however, continued to watch the filaments, and after a time noticed little dots appearing within them. These dots became more and more distinct, until finally the whole length of the organism was studded with minute ovoid bodies, which lay within the outer integument like peas within their shell. By-and-bye the integument fell to pieces, the place of the organisms being taken by a long row of seeds or spores. These observations, which were confirmed in all respects by the celebrated naturalist, Cohn of Breslau, are of the highest importance. They clear up the existing perplexity regarding the latent and visible contagia of splenic fever; for in the most conclusive manner, Koch proved the spores, as distinguished from the rods, to constitute the contagium of the fever in its most deadly and persistent form.

How did he reach this important result? Mark the answer. There was but one way open to him to test the activity of the contagium, and that was the inoculation with it of living animals. He operated upon guinea-pigs and rabbits, but the vast majority of his experiments were made upon mice. Inoculating them with the fresh blood of an animal suffering from splenic fever, they invariably died of the same disease within twenty or thirty hours after inoculation. He then sought to determine how the contagium maintained its vitality. Drying the infectious blood containing the rod-like organisms, in which, however, the spores were not developed, he found the contagium to be that which Dr. Sanderson calls 'fugitive.' It maintained its power

of infection for five weeks at the furthest. He then dried blood containing the fully developed spores, and exposed the substance to a variety of conditions. He permitted the dried blood to assume the form of dust; wetted this dust, allowed it to dry again, permitted it to remain for an indefinite time in the midst of putrefying matter, and subjected it to various other tests. After keeping the spore-charged blood which had been treated in this fashion for four years, he inoculated a number of mice with it, and found its action as fatal as that of blood fresh from the veins of an animal suffering from splenic fever. There was no single escape from death after inoculation by this deadly contagium. Uncounted millions of these spores are developed in the body of every animal which has died of splenic fever, and every spore of these millions is competent to produce the disease. The name of this formidable parasite is *Bacillus anthracis*.[1]

Now the very first step towards the extirpation of these contagia is the knowledge of their nature; and the knowledge brought to us by Dr. Koch will render as certain the stamping out of splenic fever as the stoppage of the plague of *pébrine* by the researches of Pasteur.[2] One small item of statistics will show what

[1] Koch found that to produce its characteristic effects the contagium of splenic fever must enter the blood; the virulently infective spleen of a diseased animal may be eaten with impunity by mice. On the other hand, the disease refuses to be communicated by inoculation to dogs, partridges, or sparrows. In their blood *Bacillus anthracis* ceases to act as a ferment. Pasteur announced more than six years ago the propagation of the vibrios of the silkworm disease called *flacherie*, both by fission and by spores. He also made some remarkable experiments on the permanence of the contagium in the form of spores. See 'Etudes sur la Maladie des Vers à Soie,' pp. 168 and 256.

[2] Surmising that the immunity enjoyed by birds might arise from the heat of their blood, which destroyed the *bacillus*, Pasteur

this implies. In the single district of Novgorod in Russia, between the years 1867 and 1870, over fifty-six thousand cases of death by splenic fever, among horses, cows, and sheep were recorded. Nor did its ravages confine themselves to the animal world, for during the time and in the district referred to, five hundred and twenty-eight human beings perished in the agonies of the same disease.

A description of the fever will help you to come to a right decision on the point which I wish to submit to your consideration. 'An animal,' says Dr. Burdon Sanderson, 'which perhaps for the previous day has declined food and shown signs of general disturbance, begins to shudder and to have twitches of the muscles of the back, and soon after becomes weak and listless. In the meantime the respiration becomes frequent and often difficult, and the temperature rises three or four degrees above the normal; but soon convulsions, affecting chiefly the muscles of the back and loins, usher in the final collapse, of which the progress is marked by the loss of all power of moving the trunk or extremities, diminution of temperature, mucous and sanguinolent alvine evacuations, and similar discharges from the mouth and nose.' In a single district of Russia, as above remarked, fifty-six thousand horses, cows, and sheep, and five hundred and twenty-eight men and women, perished in this way during a period of two or three years. What the annual fatality is throughout Europe I have no means of knowing. Doubtless it must be very great. The question, then, which I wish to submit to your judgment is this:—Is

lowered their temperature artificially, inoculated them, and *killed them*. He also raised the temperature of guinea-pigs after inoculation, and *saved them*. It is needless to dwell for a moment on the importance of this experiment.

the knowledge which reveals to us the nature, and which assures the extirpation, of a disorder so virulent and so vile, worth the price paid for it? It is exceedingly important that assemblies like the present should see clearly the issues at stake in such questions as this, and that the properly informed sense of the community should temper, if not restrain, the rashness of those who, meaning to be tender, become agents of cruelty by the imposition of short-sighted restrictions upon physiological investigations. It is a modern instance of zeal for God, but not according to knowledge, the excesses of which must be corrected by an instructed public opinion.

And now let us cast a backward glance on the field we have traversed, and try to extract from our labours such further profit as they can yield. For more than two thousand years the attraction of light bodies by amber was the sum of human knowledge regarding electricity, and for more than two thousand years fermentation was effected without any knowledge of its cause. In science one discovery grows out of another, and cannot appear without its proper antecedent. Thus, before fermentation could be understood, the microscope had to be invented, and brought to a considerable degree of perfection. Note the growth of knowledge. Leeuwenhoek, in 1680, found yeast to be a mass of floating globules, but he had no notion that the globules were alive. This was proved in 1835 by Cagniard de la Tour and Schwann. Then came the question as to the origin of such microscopic organisms, and in this connexion the memoir of Pasteur, published in the 'Annales de Chimie' for 1862, is the inauguration of a new epoch.

On that investigation all Pasteur's subsequent

labours were based. Ravages had over and over again occurred among French wines. There was no guarantee that they would not become acid or bitter, particularly when exported. The commerce in wines was thus restricted, and disastrous losses were often inflicted on the wine-grower. Every one of these diseases was traced to the life of an organism. Pasteur ascertained the temperature which killed these ferments of disease, proving it to be so low as to be perfectly harmless to the wine. By the simple expedient of heating the wine to a temperature of fifty degrees Centigrade, he rendered it inalterable, and thus saved his country the loss of millions. He then went on to vinegar—*vin aigre*, acid wine—which he proved to be produced by a fermentation set up by a little fungus called *Mycoderma aceti*. *Torula*, in fact, converts the grape juice into alcohol, and *Mycoderma aceti* converts the alcohol into vinegar. Here also frequent failures occurred, and severe losses were sustained. Through the operation of unknown causes, the vinegar often became unfit for use, sometimes indeed falling into utter putridity. It had been long known that mere exposure to the air was sufficient to destroy it. Pasteur studied all these changes, traced them to their living causes, and showed that the permanent health of the vinegar was ensured by the destruction of this life. He passed from the diseases of vinegar to the study of a malady which a dozen years ago had all but ruined the silk husbandry of France. This plague, which received the name of *pébrine*, was the product of a parasite which first took possession of the intestinal canal of the silk-worm, spread throughout its body, and filled the sack which ought to contain the viscid matter of the silk. Thus smitten, the worm would go automatically through the process of spinning, when it had nothing to spin.

Pasteur followed this parasitic destroyer from year to year, and led by his singular power of combining facts with the logic of facts, discovered eventually the precise phase in the development of the insect when the disease which assailed it could with certainty be stamped out. Pasteur's devotion to this inquiry cost him dear. He restored to France her silk husbandry, rescued thousands of her population from ruin, set the looms of Italy also to work, but emerged from his labours with one of his sides permanently paralyzed. His last investigation is embodied in a work entitled 'Studies on Beer,' in which he describes a method of rendering beer permanently unchangeable. That method is not so simple as those found effectual with wine and vinegar, but the principles which it involves are sure to receive extensive application at some future day.

There are other reflections connected with this subject which, even were they now passed over without remark, would sooner or later occur to every thoughtful mind in this assembly. I have spoken of the floating dust of the air, of the means of rendering it visible, and of the perfect immunity from putrefaction which accompanies the contact of germless infusions and moteless air. Consider the woes which these wafted particles, during historic and pre-historic ages, have inflicted on mankind; consider the loss of life in hospitals from putrefying wounds; consider the loss in places where there are plenty of wounds, but no hospitals, and in the ages before hospitals were anywhere founded; consider the slaughter which has hitherto followed that of the battle-field, when those bacterial destroyers are let loose, often producing a mortality far greater than that of the battle itself; add to this the other conception that in times of epidemic disease the self-same floating matter

has mingled with it the special germs which produce the epidemic, being thus enabled to sow pestilence and death over nations and continents—consider all this, and you will come with me to the conclusion that all the havoc of war, ten times multiplied, would be evanescent if compared with the ravages due to atmospheric dust.

This preventible destruction is going on to-day, and it has been permitted to go on for ages, without a whisper of information regarding its cause being vouchsafed to the suffering sentient world. We have been scourged by invisible thongs, attacked from impenetrable ambuscades, and it is only to-day that the light of science is being let in upon the murderous dominion of our foes. Facts like these excite in me the thought that the rule and governance of this universe are different from what we in our youth supposed them to be—that the inscrutable Power, at once terrible and beneficent, in whom we live and move and have our being and our end, is to be propitiated by means different from those usually resorted to. The first requisite towards such propitiation is *knowledge*; the second is *action*, shaped and illuminated by that knowledge. Of knowledge we already see the dawn, which will open out by-and-by to perfect day; while the action which is to follow has its unfailing source and stimulus in the moral and emotional nature of man—in his desire for personal well-being, in his sense of duty, in his compassionate sympathy with the sufferings of his fellow-men. 'How often,' says Dr. William Budd in his celebrated work on Typhoid Fever,—'How often have I seen in past days, in the single narrow chamber of the day-labourer's cottage the father in the coffin, the mother in the sick-bed in muttering delirium, and nothing to relieve the desolation of the children but the devotion of some poor neigh-

bour, who in too many cases paid the penalty of her kindness in becoming herself the victim of the same disorder!' From the vantage ground already won I look forward with confident hope to the triumph of medical art over scenes of misery like that here described. The cause of the calamity being once clearly revealed, not only to the physician, but to the public, whose intelligent co-operation is absolutely essential to success, the final victory of humanity is only a question of time. We have already a foretaste of that victory in the triumphs of surgery as practised at your doors.

SPONTANEOUS GENERATION.[1]

V.

WITHIN ten minutes' walk of a little cottage which I have recently built in the Alps, there is a small lake, fed by the melted snows of the upper mountains. During the early weeks of summer no trace of life is to be discerned in this water; but invariably towards the end of July, or the beginning of August, swarms of tailed organisms are seen enjoying the sun's warmth along the shallow margins of the lake, and rushing with audible patter into deeper water at the approach of danger. The origin of this periodic crowd of living things is by no means obvious. For years I had never noticed in the lake either an adult frog, or the smallest fragment of frog spawn; so that were I not otherwise informed, I should have found the conclusion of Mathiole a natural one, namely, that tadpoles are generated in lake mud by the vivifying action of the sun.

The checks which experience alone can furnish being absent, the spontaneous generation of creatures quite as high as the frog in the scale of being was assumed for ages to be a fact. Here, as elsewhere, the dominant mind of Aristotle stamped its notions on the world at large. For nearly twenty centuries after him men found no difficulty in believing in cases of

[1] 'The Nineteenth Century,' January 1878.

spontaneous generation which would now be rejected as monstrous by the most fanatical supporter of the doctrine. Shell-fish of all kinds were considered to be without parental origin. Eels were supposed to spring spontaneously from the fat ooze of the Nile. Caterpillars were the spontaneous products of the leaves on which they fed; while winged insects, serpents, rats, and mice were all thought capable of being generated without sexual intervention.

The most copious source of this life without an ancestry was putrefying flesh; and, lacking the checks imposed by fuller investigation, the conclusion that flesh possesses and exerts this generative power is a natural one. I well remember when a child of ten or twelve seeing a joint of imperfectly salted beef cut into, and coils of maggots laid bare within the mass. Without a moment's hesitation I jumped to the conclusion that these maggots had been spontaneously generated in the meat. I had no knowledge which could qualify or oppose this conclusion, and for the time it was irresistible. The childhood of the individual typifies that of the race, and the belief here enunciated was that of the world for nearly two thousand years.

To the examination of this very point the celebrated Francesco Redi, physician to the Grand Dukes Ferdinand II. and Cosmo III. of Tuscany, and a member of the Academy del Cimento, addressed himself in 1668. He had seen the maggots of putrefying flesh, and reflected on their possible origin. But he was not content with mere reflection, nor with the theoretic guesswork which his predecessors had founded upon their imperfect observations. Watching meat during its passage from freshness to decay, prior to the appearance of maggots he invariably observed flies

buzzing round the meat and frequently alighting on it. The maggots, he thought, might be the half-developed progeny of these flies.

The inductive guess precedes experiment, by which, however, it must be finally tested. Redi knew this, and acted accordingly. Placing fresh meat in a jar and covering the mouth with paper, he found that, though the meat putrefied in the ordinary way, it never bred maggots, while the same meat placed in open jars soon swarmed with these organisms. For the paper cover he then substituted fine gauze, through which the odour of the meat could rise. Over it the flies buzzed, and on it they laid their eggs, but, the meshes being too small to permit the eggs to fall through, no maggots were generated in the meat. They were, on the contrary, hatched upon the gauze. By a series of such experiments Redi destroyed the belief in the spontaneous generation of maggots in meat, and with it doubtless many related beliefs. The combat was continued by Vallisneri, Schwammerdam, and Réaumur, who succeeded in banishing the notion of spontaneous generation from the scientific minds of their day. Indeed, as regards such complex organisms as those which formed the subject of their researches, the notion was banished for ever.

But the discovery and improvement of the microscope, though giving a death-blow to much that had been previously written and believed regarding spontaneous generation, brought also into view a world of life formed of individuals so minute—so close as it seemed to the ultimate particles of matter—as to suggest an easy passage from atoms to organisms. Animal and vegetable infusions exposed to the air were found clouded and crowded with creatures far beyond the reach of unaided vision, but perfectly visible to an eye

strengthened by the microscope. With reference to their origin these organisms were called 'Infusoria.' Stagnant pools were found full of them, and the obvious difficulty of assigning a germinal origin to existences so minute furnished the precise condition necessary to give new play to the notion of heterogenesis or spontaneous generation.

The scientific world was soon divided into two hostile camps, the leaders of which only can here be briefly alluded to. On the one side, we have Buffon and Needham, the former postulating his 'organic molecules,' and the latter assuming the existence of a special 'vegetative force' which drew the molecules together so as to form living things. On the other side, we have the celebrated Abbé Lazzaro Spallanzani, who in 1777 published results counter to those announced by Needham in 1748. They were obtained by methods so precise as to completely overthrow the convictions based upon the labours of his predecessor. Charging his flasks with organic infusions, he sealed their necks with the blowpipe, subjected them in this condition to the heat of boiling water, and subsequently exposed them to temperatures favourable to the development of life. The infusions continued unchanged for months, and when the flasks were subsequently opened no trace of life was found.

Here I may forestall matters so far as to say that the success of Spallanzani's experiments depended wholly on the locality in which he worked. The air around him must have been free from the more obdurate infusorial germs, for otherwise the process he followed would, as was long afterwards proved by Wyman, have infallibly yielded life. But his refutation of the doctrine of spontaneous generation is not the less valid on this account. Nor is it in any way upset by the fact,

that others in repeating his experiments obtained life where he obtained none. Rather is the refutation strengthened by such differences. Given two experimenters equally skilful and equally careful, operating in different places on the same infusion, in the same way, and assuming the one to obtain life while the other fails to obtain it; then its well-established absence in the one case proves that some ingredient foreign to the infusion must be its cause in the other.

Spallanzani's sealed flasks contained but small quantities of air, and as oxygen was afterwards shown to be generally essential to life, it was thought that the absence of life observed by Spallanzani might have been due to the lack of this vitalizing gas. To dissipate this doubt, Schulze in 1836 half filled a flask with distilled water to which animal and vegetable matters were added. First boiling his infusion to destroy whatever life it might contain, Schulze sucked daily into his flask air which had passed through a series of bulbs containing concentrated sulphuric acid, where all germs of life suspended in the air were supposed to be destroyed. From May to August this process was continued without any development of infusorial life.

Here again the success of Schulze was due to his working in comparatively pure air, but even in such air his experiment is a very risky one. Germs will pass unwetted and unscathed through sulphuric acid unless the most special care is taken to detain them. I have repeatedly failed, by repeating Schulze's experiments, to obtain his results. Others have failed likewise. The air passes in bubbles through the bulbs, and to render the method secure, the passage of the air must be so slow as to cause the whole of its floating matter, even to the very core of each bubble, to touch the surrounding liquid. But if this precaution be observed,

water will be found quite as effectual as sulphuric acid. By the aid of an air-pump, in an highly infective atmosphere I have thus drawn air for weeks without intermission, first through bulbs containing water, and afterwards through vessels containing organic infusions, without any appearance of life. The germs were not killed by the water, but they were effectually intercepted, while the objection that the air had been injured by being brought into contact with strongly corrosive substances was annulled.

The brief paper of Schulze, published in Poggendorf's *Annalen* for 1836, was followed in 1837 by another short and pregnant communication by Schwann. Redi, as we have seen, traced the maggots of putrefying flesh to the eggs of flies. But he did not and he could not know the meaning of putrefaction itself. He had not the instrumental means to inform him that *it* also is a phenomenon attendant on the development of life. This was first proved in the paper now alluded to. Schwann placed flesh in a flask filled to one-third of its capacity with water, sterilized the flask by boiling, and then supplied it for months with calcined air. Throughout this time there appeared no mould, no infusoria, no putrefaction; the flesh remained unaltered, while the liquid continued as clear as it was immediately after boiling. Schwann then varied his experimental argument, with no alteration in the result. His final conclusion was, that putrefaction is due to decompositions of organic matter attendant on the multiplication therein of minute organisms. These organisms were derived not from the air, but from something contained in the air, which was destroyed by a sufficiently high temperature. There never was a more determined opponent of the doctrine of spontaneous generation than Schwann, though a strange attempt was made a year and a half

ago to enlist him and others equally opposed to it on the side of the doctrine.

The physical character of the agent which produces putrefaction was further revealed by Helmholtz in 1843. By means of a membrane he separated a sterilized putrescible liquid from a putrefying one. The sterilized infusion remained perfectly intact. Hence it was not the liquid of the putrefying mass—for that could freely diffuse through the membrane—but something contained in the liquid, and which was stopped by the membrane, that caused the putrefaction. In 1854 Schroeder and Von Dusch struck into this inquiry, which was subsequently followed up by Schroeder alone. These able experimenters employed plugs of cotton-wool to filter the air supplied to their infusions. Fed with such air, in the great majority of cases the putrescible liquids remained perfectly sweet after boiling. Milk formed a conspicuous exception to the general rule. It putrefied after boiling, though supplied with carefully filtered air. The researches of Schroeder bring us up to the year 1859.

In that year a book was published which seemed to overturn some of the best established facts of previous investigators. Its title was *Hétérogénie*, and its author was F. A. Pouchet, Director of the Museum of Natural History at Rouen. Ardent, laborious, learned, full not only of scientific but of metaphysical fervour, he threw his whole energy into the inquiry. Never did a subject require the exercise of the cold critical faculty more than this one—calm study in the unravelling of complex phenomena, care in the preparation of experiments, care in their execution, skilful variation of conditions, and incessant questioning of results until repetition had placed them beyond doubt or question. To a man of Pouchet's temperament the subject was full of danger

—danger not lessened by the theoretic bias with which he approached it. This is revealed by the opening words of his preface: 'Lorsque, par la méditation, il fut évident pour moi que la génération spontanée était encore l'un des moyens qu'emploie la nature pour la reproduction des êtres, je m'appliquai à découvrir par quels procédés on pouvait parvenir à en mettre les phénomènes en évidence.' It is needless to say that such a prepossession required a strong curb. Pouchet repeated the experiments of Schulze and Schwann with results diametrically opposed to theirs. He heaped experiment upon experiment and argument upon argument, spicing with the sarcasm of the advocate the logic of the man of science. In view of the multitudes required to produce the observed results, he ridiculed the assumption of atmospheric germs. This was one of his strongest points. 'Si les Proto-organismes que nous voyons pulluler partout et dans tout, avaient leurs germes disséminés dans l'atmosphère, dans la proportion mathématiquement indispensable à cet effet, l'air en serait totalement obscurci, car ils devraient s'y trouver beaucoup plus serrés que les globules d'eau qui forment nos nuages épais. Il n'y a pas là la moindre exagération.' Recurring to the subject, he exclaims: 'L'air dans lequel nous vivons aurait presque la densité du fer.' There is often a virulent contagion in a confident tone, and this hardihood of argumentative assertion was sure to influence minds swayed not by knowledge, but by authority. Had Pouchet known that 'the blue ethereal sky' is formed of suspended particles, through which the sun freely shines, he would hardly have ventured upon this line of argument.

Pouchet's pursuit of this inquiry strengthened the conviction with which he began it, and landed him in downright credulity in the end. I do not question

his ability as an observer, but the inquiry needed a disciplined experimenter. This latter implies not mere ability to look at things as Nature offers them to our inspection, but to force her to show herself under conditions prescribed by the experimenter himself. Here Pouchet lacked the necessary discipline. Yet the vigour of his onset raised clouds of doubt, which for a time obscured the whole field of inquiry. So difficult indeed did the subject seem, and so incapable of definite solution, that when Pasteur made known his intention to take it up, his friends Biot and Dumas expressed their regret, earnestly exhorting him to set a definite and rigid limit to the time he purposed spending in this apparently unprofitable field.[1]

Schooled by his education as a chemist, and by special researches on the closely related question of fermentation, Pasteur took up this subject under particularly favourable conditions. His work and his culture had given strength and finish to his natural aptitudes. In 1862, accordingly, he published a paper 'On the Organized Corpuscles existing in the Atmosphere,' which must for ever remain classical. By the most ingenious devices he collected the floating particles of the air surrounding his laboratory in the Rue d'Ulm, and subjected them to microscopic examination. Many of them he found to be organized particles. Sowing them in sterilized infusions, he obtained abundant crops of microscopic organisms. By more refined methods he repeated and confirmed the experiments of Schwann, which had been contested by Pouchet,

[1] 'Je ne conseillerais à personne,' said Dumas to his already famous pupil, 'de rester trop longtemps dans ce sujet.'—*Annales de Chimie et de Physique*, 1862, vol. lxiv. p. 22. Since that time the illustrious Perpetual Secretary of the Academy of Sciences has had good reason to revise this 'counsel.'

Montegazza, Joly, and Musset. He also confirmed the experiments of Schroeder and Von Dusch. He showed that the cause which communicated life to his infusions was not uniformly diffused through the air; that there were aërial interspaces which possessed no power to generate life. Standing on the Mer de Glace, near the Montanvert, he snipped off the ends of a number of hermetically-sealed flasks containing organic infusions. One out of twenty of the flasks thus supplied with glacier air showed signs of life afterwards, while eight out of twenty of the same infusions, supplied with the air of the plains, became crowded with life. He took his flasks into the caves under the Observatory of Paris, and found the still air in these caves devoid of generative power. These and other experiments, carried out with a severity perfectly obvious to the instructed scientific reader, and accompanied by a logic equally severe, restored the conviction that, even in these lower reaches of the scale of being, life does not appear without the operation of antecedent life.

The main position of Pasteur has been strengthened by practical researches of the most momentous kind. He has applied the knowledge won from his inquiries to the preservation of wine and beer, to the manufacture of vinegar, to the staying of the plague which threatened utter destruction of the silk husbandry of France, and to the examination of other formidable diseases which assail the higher animals, including man. His relation to the improvements which Professor Lister has introduced into surgery is shown by a letter quoted in his *Etudes sur la Bière*.[1] Professor Lister there expressly thanks Pasteur for having given him the only principle which could have conducted the antiseptic system to a successful issue. The strictures regarding defects of reasoning, to which we have been lately accustomed,

[1] P. 43.

whatever light they may shed upon their author, throw no shade upon Pasteur.

Redi, as we have seen, proved the maggots of putrefying flesh to be derived from the eggs of flies; Schwann proved putrefaction itself to be the concomitant of far lower forms of life than those dealt with by Redi. Our knowledge here, as elsewhere in connexion with this subject, has been vastly extended by Professor Cohn, of Breslau. 'No putrefaction,' he says, 'can occur in a nitrogenous substance if its bacteria be destroyed and new ones prevented from entering it. Putrefaction begins as soon as bacteria, even in the smallest numbers, are admitted either accidentally or purposely. It progresses in direct proportion to the multiplication of the bacteria, it is retarded when they exhibit low vitality, and is stopped by all influences which either hinder their development or kill them. All bactericidal media are therefore antiseptic and disinfecting.'[1] It was these organisms acting in wound and abscess which so frequently converted our hospitals into charnel-houses, and it is their destruction by the antiseptic system that now renders justifiable operations which no surgeon would have attempted a few years ago. The gain is immense—to the practising surgeon as well as to the patient practised upon. Contrast the anxiety of never feeling sure whether the most brilliant operation might not be rendered nugatory by the access of a few particles of unseen hospital dust, with the comfort derived from the knowledge that all power of mischief on the part of such dust has been surely and certainly

[1] In his last excellent memoir Cohn expresses himself thus: 'Wer noch heut die Fäulniss von einer spontanen Dissociation der Proteinmolecule, oder von einem unorganisirten Ferment ableitet, oder gar aus "Stickstoffsplittern" die Balken zur Stütze seiner Fäulnisstheorie zu zimmern versucht, hat zuerst den Satz "keine Fäulniss ohne Bacterium Termo" zu widerlegen.'

annihilated. But the action of living contagia extends beyond the domain of the surgeon. The power of reproduction and indefinite self-multiplication which is characteristic of living things, coupled with the undeviating fact of contagia 'breeding true,' has given strength and consistency to a belief long entertained by penetrating minds, that epidemic diseases generally are the concomitants of parasitic life. 'There begins to be faintly visible to us a vast and destructive laboratory of nature wherein the diseases which are most fatal to animal life, and the changes to which dead organic matter is passively liable, appear bound together by what must at least be called a very close analogy of causation.'[1] According to this view, which, as I have said, is daily gaining converts, a contagious disease may be defined as a conflict between the person smitten by it and a specific organism which multiplies at his expense, appropriating his air and moisture, disintegrating his tissues, or poisoning him by the decompositions incident to its growth.

During the ten years extending from 1859 to 1869, researches on radiant heat in its relations to the gaseous form of matter occupied my continual attention. When air was experimented on, I had to cleanse it effectually of floating matter, and while doing so I was surprised to notice that, at the ordinary rate of transfer, such matter passed freely through alkalis, acids, alcohols, and ethers. The eye being kept sensitive by darkness, a concentrated beam of light was found to be a most searching test for suspended matter both in water and in air—a test indeed indefinitely more searching and severe than that furnished by the most powerful microscope. With the aid of such a beam I examined

[1] Report of the Medical Officer of the Privy Council, 1874, p. 5.

air filtered by cotton-wool; air long kept free from agitation, so as to allow the floating matter to subside; calcined air, and air filtered by the deeper cells of the human lungs. In all cases the correspondence between my experiments and those of Schwann, Schroeder, Pasteur, and Lister in regard to spontaneous generation was perfect. The air which they found inoperative was proved by the luminous beam to be optically pure and therefore germless. Having worked at the subject both by experiment and reflection, on Friday evening, January 21, 1870, I brought it before the members of the Royal Institution. Two or three months subsequently, for sufficient practical reasons, I ventured to direct public attention to the subject in a letter to the *Times*. Such was my first contact with this important question.

This letter, I believe, gave occasion for the first public utterance of Dr. Bastian in relation to this subject. He did me the honour to inform me, as others had informed Pasteur, that the subject 'pertains to the biologist and physician.' He expressed 'amazement' at my reasoning, and warned me that before what I had done could be undone 'much irreparable mischief might be occasioned.' With far less preliminary experience to guide and warn him, the English heterogenist was far bolder than Pouchet in his experiments, and far more adventurous in his conclusions. With organic infusions he obtained the results of his celebrated predecessor, but he did much more—the atoms and molecules of inorganic liquids passing under his manipulation into those more 'complex chemical compounds,' which we dignify by calling them 'living organisms.'[1] As re-

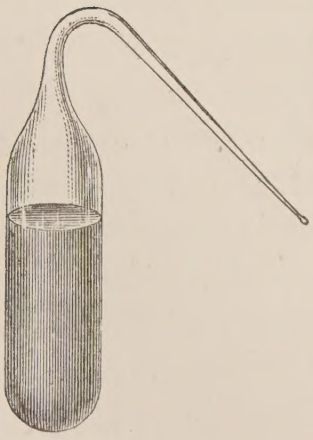
[1] It is further held that bacteria or allied organisms are prone to be engendered as correlative products, coming into existence in the several fermentations, just as independently as other less complex chemical compounds.'—Bastian, *Trans. of Pathological Society*, vol. xxvi. 258.

gards the public who take an interest in such things, and apparently also as regards a large portion of the medical profession, our extremely clever countryman succeeded in restoring the subject to a state of uncertainty similar to that which followed the publication of Pouchet's volume in 1859.

It is desirable that this uncertainty should be removed from all minds, and doubly desirable on practical grounds that it should be removed from the minds of medical men. In the present article, therefore, I propose discussing this question face to face with some eminent and fair-minded member of the medical profession who, as regards spontaneous generation, entertains views adverse to mine. Such a one it would be easy to name; but it is perhaps better to rest in the impersonal. I shall therefore simply call my proposed co-inquirer my friend. With him at my side, I shall endeavour, to the best of my ability, so to conduct this discussion that he who runs may read and that he who reads may understand.

Let us begin at the beginning. I ask my friend to step into the laboratory of the Royal Institution, where I place before him a basin of thin turnip slices barely covered with distilled water kept at a temperature of 120° Fahr. After digesting the turnip for four or five hours we pour off the liquid, boil it, filter it, and obtain an infusion as clear as filtered drinking water. We cool the infusion, test its specific gravity, and find it to be 1006 or higher—water being 1000. A number of small clean empty flasks, of the shape shown on the margin, are before us. One of

FIG. 23.



them is slightly warmed with a spirit-lamp, and its open end is then dipped into the turnip infusion. The warmed glass is afterwards chilled, the air within the flasks cools, contracts, and is followed in its contraction by the infusion. Thus we get a small quantity of liquid into the flask. We now heat this liquid carefully. Steam is produced, which issues from the open neck, carrying the air of the flask along with it. After a few seconds' ebullition, the open neck is again plunged into the infusion. The steam within the flask condenses, the liquid enters to supply its place, and in this way we fill our little flask to about four-fifths of its volume. This description is typical; we may thus fill a thousand flasks with a thousand different infusions.

I now ask my friend to notice a trough made of sheet copper, with two rows of handy little Bunsen burners underneath it. This trough, or bath, is nearly filled with oil; a piece of thin plank constitutes a kind of lid for the oil-bath. The wood is perforated with circular apertures wide enough to allow our small flask to pass through and plunge itself in the oil, which has been heated, say, to 250° Fahr. Clasped all round by the hot liquid, the infusion in the flask rises to its boiling point, which is not sensibly over 212° Fahr. Steam issues from the open neck of the flask, and the boiling is continued for five minutes. With a pair of small brass tongs, an assistant now seizes the neck near its junction with the flask, and partially lifts the latter out of the oil. The steam does not cease to issue, but its violence is abated. With a second pair of tongs held in one hand, the neck of the flask is seized close to its open end, while with the other hand a Bunsen's flame or an ordinary spirit flame is brought under the middle of the neck. The glass reddens, whitens, softens, and as it is gently drawn out the neck diminishes in dia-

meter, until the canal is completely blocked up. The second pair of tongs with the fragment of severed neck being withdrawn, the flask, with its contents diminished by evaporation, is lifted from the oil-bath perfectly sealed hermetically.

Sixty such flasks filled, boiled, and sealed in the manner described, and containing strong infusions of beef, mutton, turnip, and cucumber, are carefully packed in sawdust, and transported to the Alps. Thither, to an elevation of about 7,000 feet above the sea, I invite my co-inquirer to accompany me. It is the month of July, and the weather is favourable to putrefaction. We open our box at the Bel Alp, and count out fifty-four flasks, with their liquids as clear as filtered drinking water. In six flasks, however, the infusion is found muddy. We closely examine these, and discover that every one of them has had its fragile end broken off in the transit from London. Air has entered the flasks, and the observed muddiness is the result. My colleague knows as well as I do what this means. Examined with a pocket-lens, or even with a microscope of insufficient power, nothing living is seen in the muddy liquid; but regarded with a magnifying power of a thousand diameters or so, what an astonishing appearance does it present! Leeuwenhoek estimated the population of a single drop of stagnant water at 500,000,000: probably the population of a drop of our turbid infusion would be this many times multiplied. The field of the microscope is crowded with organisms, some wabbling slowly, others shooting rapidly across the microscopic field. They dart hither and thither like a rain of minute projectiles; they pirouette and spin so quickly round, that the retention of the retinal impression transforms the little living rod into a twirling wheel. And yet the most celebrated naturalists tells us they are vegetables.

From the rod-like shape which they so frequently assume, these organisms are called 'bacteria'—a term, be it here remarked, which covers organisms of very diverse kinds.

Has this multitudinous life been spontaneously generated in these six flasks, or is it the progeny of living germinal matter carried into the flasks by the entering air? If the infusions have a self-generative power, how are the sterility and consequent clearness of the fifty-four uninjured flasks to be accounted for? My colleague may urge—and fairly urge—that the assumption of germinal matter is by no means necessary; that the air itself may be the one thing needed to wake up the dormant infusions. We will examine this point immediately. But meanwhile I would remind him that I am working on the exact lines laid down by our most conspicuous heterogenist. He distinctly affirms that the withdrawal of the atmospheric pressure above the infusion favours the production of organisms; and he accounts for their absence in tins of preserved meat, fruit, and vegetables, by the hypothesis that fermentation *has* begun in such tins, that gases *have* been generated, the pressure of which has stifled the incipient life and stopped its further development.[1] This is the new theory of preserved meats. Had Dr. Bastian pierced a tin of preserved meat, fruit, or vegetable under water with the view of testing its truth, he would have found it erroneous. In well-preserved tins he would have found, not an outrush of gas, but an inrush of water. I have noticed this recently in tins which have lain perfectly good for sixty-three years in the Royal Institution. Modern tins, subjected to the same test, yielded the same result. From time to time, moreover, during the last two years, I have placed glass

[1] 'Beginnings of Life,' vol. i. p. 418.

tubes, containing clear infusions of turnip, hay, beef, and mutton, in iron bottles, and subjected them to air-pressures varying from ten to twenty-seven atmospheres —pressures, it is needless to say, far more than sufficient to tear a preserved meat tin to shreds. After ten days these infusions were taken from their bottles rotten with putrefaction and teeming with life. Thus collapses an hypothesis which had no rational foundation, and which could never have seen the light had any well-directed attempt been made to verify it.

Our fifty-four vacuous and pellucid flasks also declare against the heterogenist. We expose them to a warm Alpine sun by day, and at night we suspend them in a warm kitchen. Four of them have been accidentally broken; but at the end of a month we find the fifty remaining ones as clear as at the commencement. There is no sign of putrefaction or of life in any of them. We divide these flasks into two groups of twenty-three and twenty-seven respectively (an accident of counting rendered the division uneven). The question now is whether the admission of air can liberate any generative energy in the infusions. Our next experiment will answer this question and something more. We carry the flasks to a hayloft, and there, with a pair of steel pliers, snip off the sealed ends of the group of three-and-twenty. Each snipping off is of course followed by an inrush of air. We now carry our twenty-seven flasks, our pliers, and a spirit-lamp, to a ledge overlooking the Aletsch glacier, about 200 feet above the hayloft, from which ledge the mountain falls almost precipitously to the north-east for about a thousand feet. A gentle wind blows towards us from the north-east—that is, across the crests and snow-fields of the Oberland mountains. We are therefore bathed by air which must have been for a good while out of practical

contact with either animal or vegetable life. I stand carefully to leeward of the flasks, for no dust or particle from my clothes or body must be blown towards them. An assistant ignites the spirit-lamp, into the flame of which I plunge the pliers, thereby destroying all attached germs or organisms. Then I snip off the sealed end of the flask. Prior to every snipping the same process is gone through, no flask being opened without the previous cleansing of the pliers by the flame. In this way we charge our seven-and-twenty flasks with clean vivifying mountain air.

We place the fifty flasks, with their necks open, over a kitchen stove, in a temperature varying from 50° to 90° Fahr., and in three days find twenty-one out of the twenty-three flasks opened on the hayloft invaded by organisms—two only of the group remaining free from them. After three weeks' exposure to precisely the same conditions, *not one of the twenty-seven flasks opened in free air had given way.* No germ from the kitchen air had ascended the narrow necks, the flasks being shaped so as to avoid this contingency. They are still in the Alps, as clear, I doubt not, and as free from life as they were when sent off from London.[1]

What is my colleague's conclusion from the experiment before us? Twenty-seven putrescible infusions, first in vacuo, and afterwards supplied with the most invigorating air, have shown no sign of putrefaction or of life. And as to the others, I almost shrink from asking him whether the hayloft has rendered them spontaneously generative. Is not the inference here imperative that it is not the air of the loft—which is connected through a constantly open door with the general atmosphere—but something contained in the

[1] An actual experiment made at the Bel Alp is here described.

air, that has produced the effects observed? What is this something? A sunbeam entering through a chink in the roof or wall, and traversing the air of the loft, would show it to be laden with suspended dust particles. Indeed the dust is distinctly visible in the diffused daylight. Can *it* have been the origin of the observed life? If so, are we not bound by all antecedent experience to regard these fruitful particles as the germs of the life observed?

The name of Baron Liebig has been constantly mixed up with these discussions. 'We have,' it is said, 'his authority for assuming that dead decaying matter can produce fermentation.' True, but with Liebig fermentation was by no means synonymous with *life*. It meant, according to him, the shaking asunder by chemical disturbance of unstable molecules. Does the life of our flasks, then, proceed from *dead* particles? If my co-inquirer should reply 'Yes,' then I would ask him, 'What warrant does Nature offer for such an assumption? Where, amid the multitude of vital phenomena in which her operations have been clearly traced, is the slightest countenance given to the notion that the sowing of dead particles can produce a living crop?' With regard to Baron Liebig, had he studied the revelations of the microscope in relation to these questions, a mind so penetrating could never have missed the significance of the facts revealed. He, however, neglected the microscope, and fell into error—but not into error so gross as that in support of which his authority has been invoked. Were he now alive, he would, I doubt not, repudiate the use often made of his name—Liebig's view of fermentation was at least a scientific one, founded on profound conceptions of molecular instability. But this view by no means involves the notion that the planting of dead particles

—'Stickstoffsplittern,' as Cohn contemptuously calls them—is followed by the sprouting of infusorial life.

Let us now return to London and fix our attention on the dust of *its* air. Suppose a room in which the housemaid has just finished her work to be completely closed, with the exception of an aperture in a shutter through which a sunbeam enters and crosses the room. The floating dust reveals the track of the light. Let a lens be placed in the aperture to condense the beam. Its parallel rays are now converged to a cone, at the apex of which the dust is raised to almost unbroken whiteness by the intensity of its illumination. Defended from all glare, the eye is peculiarly sensitive to this scattered light. The floating dust of London rooms is organic, and may be burned without leaving visible residue. The action of a spirit-lamp flame upon the floating matter has been elsewhere thus described:—

In a cylindrical beam which strongly illuminated the dust of our laboratory, I placed an ignited spirit lamp. Mingling with the flame, and round its rim, were seen curious wreaths of darkness resembling an intensely black smoke. On placing the flame at some distance below the beam, the same dark masses stormed upwards. They were blacker than the blackest smoke ever seen issuing from the funnel of a steamer; and their resemblance to smoke was so perfect as to prompt the conclusion that the apparently pure flame of the alcohol-lamp required but a beam of sufficient intensity to reveal its clouds of liberated carbon.

But is the blackness smoke? This question presented itself in a moment, and was thus answered: A red-hot poker was placed underneath the beam; from it the black wreaths also ascended. A large hydrogen flame, which emits no smoke, was next employed, and it also produced with augmented copiousness those whirling masses of darkness. Smoke being out of the question, what is the black-

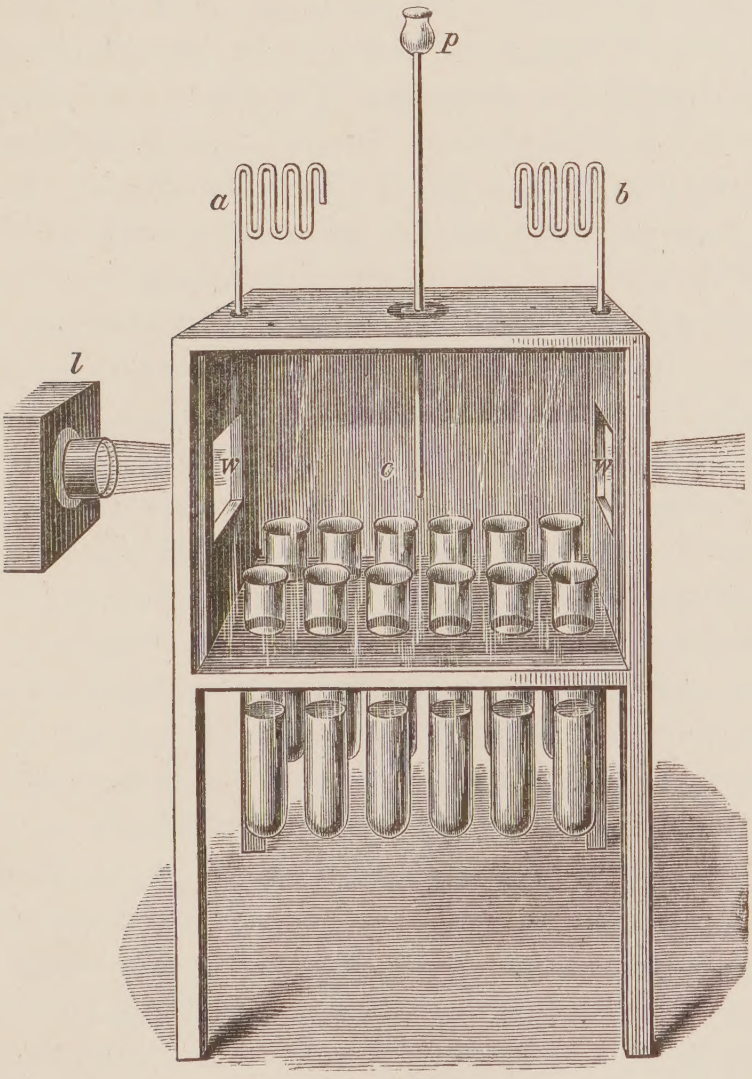
ness? It is simply that of stellar space; that is to say, blackness resulting from the absence from the track of the beam of all matter competent to scatter its light. When the flame was placed below the beam, the floating matter was destroyed *in situ;* and the heated air, freed from this matter, rose into the beam, jostled aside the illuminated particles, and substituted for their light the darkness due to its own perfect transparency. Nothing could more forcibly illustrate the invisibility of the agent which renders all things visible. The beam crossed, unseen, the black chasm formed by the transparent air, while, at both sides of the gap, the thick-strewn particles shone out like a luminous solid under the powerful illumination.[1]

Supposing an infusion intrinsically barren, but readily susceptible of putrefaction when exposed to common air, to be brought into contact with this unilluminable air, what would be the result? It would never putrefy. It might, however, be urged that the air is spoiled by its violent calcination. Oxygen passed through a spirit-lamp flame is, it may be thought, no longer the oxygen suitable for the development and maintenance of life. We have an easy escape from this difficulty, which is based, however, upon the unproved assumption that the air has been affected by the flame. Let a condensed beam be sent through a large flask or bolthead containing common air. The track of the beam is seen within the flask—the dust revealing the light, and the light revealing the dust. Cork the flask, stuff its neck with cotton-wool, or simply turn it mouth downwards and leave it undisturbed for a day or two. Examined afterwards with the luminous beam, no track is visible; the light passes through the flask as through a vacuum. The floating matter has abolished itself, being now attached to the interior surface of the flask.

[1] See pp. 3 and 4 of this volume.

Were it our object, as it will be subsequently, to effectually detain the dirt, we might coat that surface with some sticky substance. Here, then, without 'torturing' the air in any way, we have found a means of ridding

FIG. 24.



it, or rather of enabling it to rid itself, of floating matter.

We have now to devise a means of testing the action of such spontaneously purified air upon putrescible infusions. Wooden chambers, or cases, are accordingly

constructed, having glass fronts, side-windows, and back-doors. Through the bottoms of the chambers test-tubes pass air-tight; their open ends, for about one-fifth of the length of the tubes, being within the chambers. Provision is made for a free connexion through sinuous channels between the inner and the outer air. Through such channels, though open, no dust will reach the chamber. The top of each chamber is perforated by a circular hole two inches in diameter, closed air-tight by a sheet of india-rubber. This is pierced in the middle by a pin, and through the pin-hole is pushed the shank of a long pipette, ending above in a small funnel. The shank also passes through a stuffing-box of cotton-wool moistened with glycerine; so that, tightly clasped by the rubber and wool, the pipette is not likely in its motions up and down to carry any dust into the chamber. The annexed woodcut (fig. 24) shows a chamber, with six test-tubes, its side-windows *w w*, its pipette *p* c, and its sinuous channels *a b* which connect the air of the chamber with the outer air.

The chamber is carefully closed and permitted to remain quiet for two or three days. Examined at the beginning by a beam sent through its windows, the air is found laden with floating matter, which in three days has wholly disappeared. To prevent its ever rising again, the internal surface of the chamber was at the outset coated with glycerine. The fresh but putrescible liquid is introduced into the six tubes in succession by means of the pipette. Permitted to remain without further precaution, every one of the tubes would putrefy and fill itself with life. The liquid has been in contact with the dust-laden air outside by which it has been infected, and the infection must be destroyed. This is done by plunging the six tubes into a bath of heated oil and boiling the infusion. The time

requisite to destroy the infection depends wholly upon its nature. Two minutes' boiling suffices to destroy some contagia, whereas two hundred minutes' boiling fails to destroy others. After the infusion has been sterilized, the oil-bath is withdrawn, and the liquid, whose putrescibility has been in no way affected by the boiling, is abandoned to the air of the chamber.

With such chambers I tested, in the autumn and winter of 1875–6, infusions of the most various kinds, embracing natural animal liquids, the flesh and viscera of domestic animals, game, fish, and vegetables. More than fifty chambers, each with its series of infusions, were tested, many of them repeatedly. There was no shade of uncertainty in any of the results. In every instance we had, within the chamber, perfect limpidity and sweetness, which in some cases lasted for more than a year—without the chamber, with the same infusion, putridity and its characteristic smells. In no instance was the least countenance lent to the notion that an infusion deprived by heat of its inherent life, and placed in contact with air cleansed of its visibly suspended matter, has any power to generate life anew.

Remembering then the number and variety of the infusions employed, and the strictness of our adherence to the rules of preparation laid down by the heterogenists themselves; remembering that we have operated upon the very substances recommended by them as capable of furnishing, even in untrained hands, easy and decisive proofs of spontaneous generation, and that we have added to their substances many others of our own —if this pretended generative power were a reality, surely it must have manifested itself somewhere. Speaking roundly, I should say that in such closed chambers at least five hundred chances have been given to it, but it has nowhere appeared.

The argument is now to be clenched by an experiment which will remove every residue of doubt as to the ability of the infusions here employed to sustain life. We open the back doors of our sealed chambers, and permit the common air with its floating particles to have access to our tubes. For three months they have remained pellucid and sweet—flesh, fish, and vegetable extracts purer than ever cook manufactured. Three days' exposure to the dusty air suffices to render them muddy, fetid, and swarming with infusorial life. The liquids are thus proved, one and all, ready for putrefaction when the contaminating agent is applied. I invite my colleague to reflect on these facts. How will he account for the absolute immunity of a liquid exposed for months in a warm room to optically pure air, and its infallible putrefaction in a few days when exposed to dust-laden air? He must, I submit, bow to the conclusion that the dust-particles are the cause of putrefactive life. And unless he accepts the hypothesis that these particles, being dead in the air, are in the liquid miraculously kindled into living things, he must conclude that the life we have observed springs from germs or organisms diffused through the atmosphere.

The experiments with hermetically-sealed flasks have reached the number of 940. A sample group of 130 of them were laid before the Royal Society on January 13, 1876. They were utterly free from life, having been completely sterilized by three minutes' boiling. Special care had been taken that the temperatures to which the flasks were exposed should include those previously alleged to be efficient. The conditions laid down by the heterogenist were accurately copied, but there was no corroboration of his results. Stress was then laid on the question of warmth, thirty degrees

being suddenly added to the temperatures with which both of us had previously worked. Waiving all protest against the caprice thus manifested, I met this new requirement also. The sealed tubes, which had proved barren in the Royal Institution, were suspended in perforated boxes, and placed under the supervision of an intelligent assistant in the Turkish Bath in Jermyn Street. From two to six days had been allowed for the generation of organisms in hermetically-sealed tubes. Mine remained in the washing-room of the bath for nine days. Thermometers placed in the boxes, and read off twice or three times a day, showed the temperature to vary from a minimum of 101° to a maximum of 112° Fahr. At the end of nine days the infusions were as clear as at the beginning. They were then removed to a warmer position. A temperature of 115° had been mentioned as particularly favourable to spontaneous generation. For fourteen days the temperature of the Turkish Bath hovered about this point, falling once as low as 106°, reaching 116° on three occasions, 118° on one, and 119° on two. The result was quite the same as that just recorded. The higher temperatures proved perfectly incompetent to develope life.

Taking the actual experiment we have made as a basis of calculation, if our 940 flasks were opened on the hayloft of the Bel Alp, 858 of them would become filled with organisms. The escape of the remaining 82 strengthens our case, proving as it does conclusively that not in the air, nor in the infusions, nor in anything continuous diffused through the air, but in *discrete particles*, suspended in the air and nourished by the infusions, we are to seek the cause of life. Our experiment proves these particles to be in some cases so far apart on the hayloft as to permit 10 per cent. of our

flasks to take in air without contracting contamination. A quarter of a century ago Pasteur proved the cause of 'so-called spontaneous generation' to be *discontinuous*. I have already referred to his observation that 12 out of 20 flasks opened on the plains escaped infection, while 19 out of 20 flasks opened on the Mer de Glace escaped. Our own experiment at the Bel Alp is a more emphatic instance of the same kind, 90 per cent. of the flasks opened in the hayloft being smitten, while not one of those opened on the free mountain ledge was attacked.

The power of the air as regards putrefactive infection is incessantly changing through natural causes, and we are able to alter it at will. Of a number of flasks opened in 1876 in the laboratory of the Royal Institution, 42 per cent. were smitten, while 58 per cent. escaped. In 1877 the proportion in the same laboratory was 68 per cent. smitten, to 32 intact. The greater mortality, so to speak, of the infusions in 1877 was due to the presence of hay which diffused its germinal dust in the laboratory air, causing it to approximate as regards infective virulence to the air of the Alpine loft. I would ask my friend to bring his scientific penetration to bear upon all the foregoing facts. They do not prove spontaneous generation to be 'impossible.' My assertions, however, relate not to 'possibilities,' but to *proofs*, and the experiments just described do most distinctly prove the evidence on which the heterogenist relies to be written on waste paper.

My colleague will not, I am persuaded, dispute these results; but he may be disposed to urge that other able and honourable men working at the same subject have arrived at conclusions different from mine. Most freely granted; but let me here recur to the remarks already made in speaking of the experiments of Spallanzani,

to the effect that the failure of others to confirm his results by no means upsets their evidence. To fix the ideas, let us suppose that my colleague comes to the laboratory of the Royal Institution, repeats there my experiments, and obtains confirmatory results; and that he then goes to University or King's College, where, operating with the same infusions, he obtains contradictory results. Will he be disposed to conclude that the selfsame substance is barren in Albemarle Street and fruitful in Gower Street or the Strand? His Alpine experience has already made known to him the literally infinite differences existing between different samples of air as regards their capacity for putrefactive infection. And, possessing this knowledge, will he not substitute for the adventurous conclusion that an organic infusion is barren at one place and spontaneously generative at another, the more rational and obvious one that the atmospheres of the two localities which have had access to the infusion are infective in different degrees?

As regards workmanship, moreover, he will not fail to bear in mind, that *fruitfulness* may be due to errors of manipulation, while *barrenness* involves the presumption of correct experiment. It is only the careful worker that can secure the latter, while it is open to every novice to obtain the former. Barrenness is the result at which the conscientious experimenter, whatever his theoretic convictions may be, ought to aim, omitting no pains to secure it, and resorting, only when there is no escape from it, to the conclusion that the life observed comes from no source which correct experiment could neutralize or avoid.

Let us again take a definite case. Supposing my colleague to operate with the same apparent care on 100 infusions—or rather on 100 samples of the same

infusion—and that 50 of them prove fruitful and 50 barren. Are we to say that the evidence for and against heterogeny is equally balanced? There are some who would not only say this, but who would treasure up the 50 fruitful flasks as 'positive' results, and lower the evidential value of the 50 barren flasks by labelling them 'negative' results. This, as shown by Dr. William Roberts, is an exact inversion of the true order of the terms positive and negative.[1] Not such, I trust, would be the course pursued by my friend. As regards the 50 fruitful flasks he would, I doubt not, repeat the experiment with redoubled care and scrutiny, and not by one repetition only, but by many, assure himself that he had not fallen into error. Such faithful scrutiny, fully carried out, would infallibly lead him to the conclusion that here, as in all other cases, the evidence in favour of spontaneous generation crumbles in the grasp of the competent inquirer.

The botanist knows that different seeds possess different powers of resistance to heat.[2] Some are killed by a momentary exposure to the boiling temperature, while others withstand it for several hours. Most of our ordinary seeds are rapidly killed, while Pouchet made known to the Paris Academy of Sciences in 1866, that certain seeds, which had been transported in fleeces of wool from Brazil, germinated after four hours' boiling. The germs of the air vary as much among themselves as the seeds of the botanist. In some localities the diffused germs are so tender that

[1] See his truly philosophical remarks on this head in the British Medical Journal, 1876, p. 282.

[2] I am indebted to Dr. Thiselton Dyer for various illustrations of such differences. It is, however, surprising that a subject of such high scientific importance should not have been more thoroughly explored. Here the scoundrels who deal in killed seeds might be able to add to our knowledge.

boiling for five minutes, or even less, would be sure to destroy them all; in other localities the diffused germs are so obstinate, that many hours' boiling would be requisite to deprive them of their power of germination. The absence or presence of a truss of desiccated hay would produce differences as great as those here described. The greatest endurance that I have ever observed—and I believe it is the greatest on record—was a case of survival after eight hours' boiling.

As regards their power of resisting heat, the infusorial germs of our atmosphere might be classified under the following and intermediate heads:—Killed in five minutes; not killed in five minutes but killed in fifteen; not killed in fifteen minutes but killed in thirty; not killed in thirty minutes but killed in an hour; not killed in an hour but killed in two hours; not killed in two but killed in three hours; not killed in three but killed in four hours. I have had several cases of survival after four and five hours' boiling, some survivals after six, and one after eight hours' boiling. Thus far has experiment actually reached; but there is no valid warrant for fixing upon even eight hours as the extreme limit of vital resistance. Probably more extended researches (though mine have been very extensive) would reveal germs more obstinate still. It is also certain that we might begin earlier, and find germs which are destroyed by a temperature far below that of boiling water. In the presence of such facts, to speak of *a* death-point of bacteria and their germs would be unmeaning—but of this more anon.

'What present warrant,' it has been asked, 'is there for supposing that a naked, or almost naked, speck of protoplasm can withstand four, six, or eight hours' boiling?' Regarding naked specks of protoplasm I make no assertion. I know nothing about them, save

as the creatures of fancy. But I do affirm, not as a 'supposition,' nor an 'assumption,' nor a 'probable guess,' nor as 'a wild hypothesis,' but as a matter of the most undoubted fact, that the spores of the hay bacillus, when thoroughly desiccated by age, have withstood the ordeal mentioned. And I further affirm that these obdurate germs, under the guidance of the knowledge that they *are* germs, can be destroyed by five minutes' boiling, or even less. This needs explanation. The finished bacterium perishes at a temperature far below that of boiling water, and it is fair to assume that the nearer the germ is to its final sensitive condition the more readily will it succumb to heat. Seeds soften before and during germination. This premised, the simple description of the following process will suffice to make its meaning understood.

An infusion infected with the most powerfully resistent germs, but otherwise protected against the floating matters of the air, is gradually raised to its boiling-point. Such germs as have reached the soft and plastic state immediately preceding their development into bacteria are thus destroyed. The infusion is then put aside in a warm room for ten or twelve hours. If for twenty-four, we might have the liquid charged with well-developed bacteria. To anticipate this, at the end of ten or twelve hours we raise the infusion a second time to the boiling temperature, which, as before, destroys all germs then approaching their point of final development. The infusion is again put aside for ten or twelve hours, and the process of heating is repeated. We thus kill the germs *in the order of their resistance*, and finally kill the last of them. No infusion can withstand this process if it be repeated a sufficient number of times. Artichoke, cucumber, and turnip infusions, which had proved specially obstinate

when infected with the germs of desiccated hay, were completely broken down by this method of discontinuous heating, three minutes being found sufficient to accomplish what three hundred minutes' continuous boiling failed to accomplish. I applied the method, moreover, to infusions of various kinds of hay, including those most tenacious of life. Not one of them bore the ordeal. These results were clearly foreseen before they were realized, so that the germ theory fulfils the test of every true theory, that test being the power of prevision.

When 'naked or almost naked specks of protoplasm' are spoken of, the imagination is drawn upon, not the objective truth of Nature. Such words sound like the words of knowledge where knowledge is really *nil*. The possibility of a 'thin covering' is conceded by those who speak in this way. Such a covering may, however, exercise a powerful protective influence. A thin pellicle of india-rubber, for example, surrounding a pea keeps it hard in boiling water for a time sufficient to reduce an uncovered pea to a pulp. The pellicle prevents imbibition, diffusion, and the consequent disintegration. A greasy or oily surface, or even the layer of air which clings to certain bodies, would act to some extent in a similar way. 'The singular resistance of green vegetables to sterilization,' says Dr. William Roberts, 'appears to be due to some peculiarity of the surface, perhaps their smooth glistening epidermis which prevented complete wetting of their surfaces.' I pointed out in 1876 that the process by which an atmospheric germ is wetted would be an interesting subject of investigation. A dry microscope covering-glass may be caused to float on water for a year. A sewing-needle may be similarly kept floating, though its specific gravity is nearly eight times that of water.

Were it not for some specific relation between the matter of the germ and that of the liquid into which it falls, wetting would be simply impossible. Antecedent to all development there must be an interchange of matter between the germ and its environment; and this interchange must obviously depend upon the relation of the germ to its encompassing liquid. Anything that hinders this interchange retards the destruction of the germ in boiling water. In my paper, published in the 'Philosophical Transactions' for 1877, I add the following remark:—

> It is not difficult to see that the surface of a seed or germ may be so affected by desiccation and other causes as practically to prevent contact between it and the surrounding liquid. The body of a germ, moreover, may be so indurated by time and dryness as to resist powerfully the insinuation of water between its constituent molecules. It would be difficult to cause such a germ to imbibe the moisture necessary to produce the swelling and softening which precede its destruction in a liquid of high temperature.

However this may be—whatever be the state of the surface, or of the body, of the spores of *Bacillus subtilis*, they do as a matter of certainty resist, under some circumstances, exposure for hours to the heat of boiling water. No theoretic scepticism can successfully stand in the way of this fact, established as it has been by hundreds—nay thousands, of rigidly conducted experiments.

We have now to test one of the principal foundations of the doctrine of spontaneous generation as formulated in this country. With this view, I place before my friend and co-inquirer two liquids which have been kept for six months in one of our sealed chambers, exposed to optically pure air. The one is a

mineral solution containing in proper proportions all the substances which enter into the composition of bacteria, the other is an infusion of turnip—it might be any one of a hundred other infusions, animal or vegetable. Both liquids are as clear as distilled water, and there is no trace of life in either of them. They are, in fact, completely sterilized. A mutton-chop, over which a little water has been poured to keep its juices from drying up, has lain for three days upon a plate in our warm room. It smells offensively. Placing a drop of the fetid mutton-juice under a microscope, it is found swarming with the bacteria of putrefaction. With a speck of the swarming liquid I inoculate the clear mineral solution and the clear turnip infusion, as a surgeon might inoculate an infant with vaccine lymph. In four-and-twenty hours the transparent liquids have become turbid throughout, and instead of being barren as at first, they are teeming with life. The experiment may be repeated a thousand times with the same invariable result. To the naked eye the liquids at the beginning were alike, being both equally transparent—to the naked eye they are alike at the end, being both equally muddy. Instead of putrid mutton-juice, we might take as a source of infection any one of a hundred other putrid liquids, animal or vegetable. So long as the liquid contains living bacteria, a speck of it communicated either to the clear mineral solution, or to the clear turnip infusion, produces in twenty-four hours the effect here described.

We now vary the experiment thus:—Opening the back-door of another closed chamber which has contained for months the pure mineral solution and the pure turnip infusion side by side, I drop into each of them a small pinch of laboratory dust. The effect here is tardier than when the speck of putrid liquid was

employed. In three days, however, after its infection with the dust, the turnip infusion is muddy, and swarming as before with bacteria. But what about the mineral solution which, in our first experiment, behaved in a manner undistinguishable from the turnip-juice? At the end of three days there is not a bacterium to be found in it. At the end of three weeks it is equally innocent of bacterial life. We may repeat the experiment with the solution and the infusion a hundred times with the same invariable result. Always in the case of the latter the sowing of the atmospheric dust yields a crop of bacteria—never in the former does the dry germinal matter kindle into active life.[1] What is the inference which the reflecting mind must draw from this experiment? Is it not as clear as day that while both liquids are able to feed the bacteria and to enable them to increase and multiply, *after they have been once fully developed*, only one of the liquids is able to develope into active bacteria the germinal dust of the air?

I invite my friend to reflect upon this conclusion; he will, I think, see that there is no escape from it. He may, if he prefers, hold the opinion, which I consider erroneous, that bacteria exist in the air, not as germs but as desiccated organisms. The inference remains, that while the one liquid is able to force the passage from the inactive to the active state, the other is not.

But this is not at all the inference which has been drawn from experiments with the mineral solution. Seeing its ability to nourish bacteria when once inoculated with the living active organism, and observing

[1] This is the deportment of the mineral solution as described by others. My own experiments would lead me to say that the development of the bacteria, though exceedingly slow and difficult, is not impossible.

that no bacteria appeared in the solution after long exposure to the air, the inference was drawn that *neither bacteria nor their germs existed in the air.* Throughout Germany the ablest literature of the subject, even that opposed to heterogeny, is infected with this error; while heterogenists at home and abroad have based upon it a triumphant demonstration of their doctrine. It is proved, they say, by the deportment of the mineral solution that neither bacteria nor their germs exist in the air; hence, if, on exposing a thoroughly sterilized turnip infusion to the air, bacteria appear, they must of necessity have been spontaneously generated. In the words of Dr. Bastian: 'We can only infer that whilst the boiled saline solution is quite incapable of engendering bacteria, such organisms are able to arise *de novo* in the boiled organic infusion.'[1]

I would ask my eminent colleague what he thinks of this reasoning now? The *datum* is—'A mineral solution exposed to common air does not develope bacteria;' the *inference* is—'Therefore if a turnip infusion similarly exposed develope bacteria, they must be spontaneously generated.' The inference, on the face of it, is an unwarranted one. But while as matter of logic it is inconclusive, as matter of fact it is chimerical. London air is as surely charged with the germs of bacteria as London chimneys are with smoke. The inference just referred to is completely disposed of by the simple question: 'Why, when your sterilized organic infusion is exposed to optically pure air, should this generation of life *de novo* utterly cease? Why should I be able to preserve my turnip-juice side by side with your saline solution for the three hundred and sixty-five days of the year, in free connexion with the general atmosphere, on the sole condition that the

[1] Proceedings of the Royal Society, vol. xxi. p. 130.

portion of that atmosphere in contact with the juice shall be visibly free from floating dust, while three days' exposure to that dust fills it with bacteria?' Am I over-sanguine in hoping that as regards the argument here set forth he who runs may read, and he who reads may understand?

We now proceed to the calm and thorough consideration of another subject, more important if possible than the foregoing one, but like it somewhat difficult to seize by reason of the very opulence of the phraseology, logical and rhetorical, in which it has been set forth. The subject now to be considered relates to what has been called 'the death-point of bacteria.' Those who happen to be acquainted with the modern English literature of the question will remember how challenge after challenge has been issued to pansspermatists in general, and to one or two home workers in particular, to come to close quarters on this cardinal point. It is obviously the stronghold of the English heterogenist. 'Water,' he says, 'is boiling merrily over a fire when some luckless person upsets the vessel so that the heated fluid exercises its scathing influence upon an uncovered portion of the body—hand, arm, or face. Here, at all events, there is no room for doubt. Boiling water unquestionably exercises a most pernicious and rapidly destructive effect upon the living matter of which we are composed.'[1] And lest it should be supposed that it is the high organization which, in this case, renders the body susceptible to heat, he refers to the action of boiling water on the hen's egg to dissipate the notion. 'The conclusion,' he says, 'would seem to force itself upon us that there is something intrinsically deleterious in the action of boiling water upon living matter—whether this matter be of high or of low organisation.'[2] Again, at another

[1] Bastian, 'Evolution,' p. 133. [2] Ibid. p. 135.

place: 'It has been shown that the briefest exposure to the influence of boiling water is destructive of all living matter.'[1]

The experiments already recorded plainly show that there is a marked difference between the dry bacterial matter of the air, and the wet, soft, and active bacteria of putrefying organic liquids. The one can be luxuriantly bred in the saline solution, the others refuse to be born there, while both of them are copiously developed in a sterilized turnip infusion. Inferences, as we have already seen, founded on the deportment of the one liquid cannot with the warrant of scientific logic be extended to the other. But this is exactly what the heterogenist has done, thus repeating as regards the death-point of bacteria the error into which he fell concerning the germs of the air. Let us boil our muddy mineral solution with its swarming bacteria for five minutes. In the soft succulent condition in which they exist in the solution not one of them escapes destruction. The same is true of the turnip infusion if it be inoculated with the living bacteria only—the aërial dust being carefully excluded. In both cases the dead organisms sink to the bottom of the liquid, and without re-inoculation no fresh organisms will arise. But the case is entirely different when we inoculate our turnip infusion with the desiccated germinal matter afloat in the air.

The 'death-point' of bacteria is the maximum temperature at which they can live, or the minimum temperature at which they cease to live. If, for example, they survive a temperature of 140°, and do not survive a temperature of 150°, the death-point lies somewhere between these two temperatures. Vaccine lymph, for example, is proved by Messrs. Braidwood and Vacher to be deprived of its power of infection by

[1] Bastian, 'Evolution,' p. 46.

brief exposure to a temperature between 140° and 150° Fahr. This may be regarded as the death-point of the lymph, or rather of the particles diffused in the lymph, which constitute the real contagium. If no time, however, be named for the application of the heat, the term 'death-point' is a vague one. An infusion, for example, which will resist five hours' continuous exposure to the boiling temperature, will succumb to five days' exposure to a temperature 50° Fahr. below that of boiling. The fully developed soft bacteria of putrefying liquids are not only killed by five minutes' boiling, but by less than a single minute's boiling—indeed, they are slain at about the same temperature as the vaccine. The same is true of the plastic, active bacteria of the turnip infusion.[1]

But, instead of choosing a putrefying liquid for inoculation, let us prepare and employ our inoculating substance in the following simple way:—Let a small wisp of hay, desiccated by age, be washed in a glass of water, and let a perfectly sterilized turnip infusion be inoculated with the washing liquid. After three hours' continuous boiling the infusion thus infected will often develope luxuriant bacterial life. Precisely the same occurs if a turnip infusion be prepared in an atmosphere well charged with desiccated hay-germs. The infusion in this case infects itself without special inoculation, and its subsequent resistance to sterilization is often very great. On the 1st of March last I purposely infected the air of our laboratory with the germinal dust of a sapless kind of hay mown in 1875. Ten groups

[1] In my paper in the Philosophical Transactions for 1876, I pointed out and illustrated experimentally the difference, as regards rapidity of development, between water-germs and air-germs; the growth from the already softened water-germs proving to be practically as rapid as from developed bacteria. This preparedness of the germ for rapid development is associated with its preparedness for rapid destruction.

of flasks were charged with turnip infusion prepared in the infected laboratory, and were afterwards subjected to the boiling temperature for periods varying from 15 minutes to 240 minutes. Out of the ten groups only one was sterilized—that, namely, which had been boiled for four hours. Every flask of the nine groups which had been boiled for 15, 30, 45, 60, 75, 90, 105, 120, and 180 minutes, respectively, bred organisms afterwards. The same is true of other vegetable infusions. On the 28th of February last, for example, I boiled six flasks, containing cucumber infusion prepared in an infected atmosphere, for periods of 15, 30, 45, 60, 120, and 180 minutes. Every flask of the group subsequently developed organisms. On the same day, in the case of three flasks, the boiling was prolonged to 240, 300, and 360 minutes; and these three flasks were completely sterilized. Animal infusions, which under ordinary circumstances are rendered infallibly barren by five minutes' boiling, behave like the vegetable infusions in an atmosphere infected with hay-germs. On the 30th of March, for example, five flasks were charged with a clear infusion of beef and boiled for 60 minutes, 120 minutes, 180 minutes, 240 minutes, and 300 minutes respectively. Every one of them became subsequently crowded with organisms, and the same happened to a perfectly pellucid mutton infusion prepared at the same time. The cases are to be numbered by hundreds in which similar powers of resistance were manifested by infusions of the most diverse kinds.

In the presence of such facts I would ask my colleague whether it is necessary to dwell for a single instant on the one-sidedness of the evidence which led to the conclusion that all living matter has its life destroyed by 'the briefest exposure to the influence of boiling water.' An infusion proved to be barren by

six months' exposure to moteless air maintained at a temperature of 90° Fahr., when inoculated with full-grown active bacteria, fills itself in two days with organisms so sensitive as to be killed by a few minutes' exposure to a temperature much below that of boiling water. But the extension of this result to the desiccated germinal matter of the air is without warrant or justification. This is obvious without going beyond the argument itself. But we have gone far beyond the argument, and proved by multiplied experiment the alleged destruction of all living matter by the briefest exposure to the influence of boiling water to be a delusion. The whole logical edifice raised upon this basis falls therefore to the ground; and the argument that bacteria and their germs, being destroyed at 140°, must, if they appear after exposure to 212°, be spontaneously generated, is, I trust, silenced for ever.

Through the precautions, variations, and repetitions observed and executed with the view of rendering its results secure, the separate vessels employed in this inquiry have mounted up in two years *to nearly ten thousand.*

Besides the philosophic interest attaching to the problem of life's origin, which will be always immense, there are the practical interests involved in the application of the doctrines here discussed to surgery and medicine. The antiseptic system, at which I have already glanced, illustrates the manner in which beneficent results of the gravest moment follow in the wake of clear theoretic insight. Surgery was once a noble art; it is now, as well, a noble science. Prior to the introduction of the antiseptic system, the thoughtful surgeon could not have failed to learn empirically that there was something in the air which often defeated the most consummate operative skill. That something the

antiseptic treatment destroys or renders innocuous. At King's College Mr. Lister operates and dresses while a fine shower of mixed carbolic acid and water, produced in the simplest manner, falls upon the wound, the lint and gauze employed in the subsequent dressing being duly saturated with the antiseptic. At St. Bartholomew's Mr. Callender employs the dilute carbolic acid without the spray; but, as regards the real point aimed at—the preventing of the wound from becoming a nidus for the propagation of septic bacteria—the practice in both hospitals is the same. Commending itself as it does to the scientifically trained mind, the antiseptic system has struck deep root in Germany.

Had space allowed, it would have given me pleasure to point out the present position of the 'germ theory' in reference to the phenomena of infectious disease, distinguishing arguments based on analogy—which, however, are terribly strong—from those based on actual observation. I should have liked to follow up the account I have already given[1] of the truly excellent researches of a young and an unknown German physician named Koch, on splenic fever, by an account of what Pasteur has recently done with reference to the same subject. Here we have before us a living contagium of the most deadly power, which we can follow from the beginning to the end of its life cycle.[2] We find it in the blood or spleen of a smitten animal in the state say of short motionless rods. When these rods are placed in a nutritive liquid on the warm stage of the microscope, we soon see them lengthening into filaments which lie, in some cases, side by side, forming

[1] 'Fortnightly Review,' November, 1876; see preceding Article on 'Fermentation.'

[2] Dallinger and Drysdale had previously shown what skill and patience can accomplish, by their admirable observations on the life history of the monads.

in others graceful loops, or becoming coiled into knots of a complexity not to be unravelled. We finally see those filaments resolving themselves into innumerable spores, each with death potentially housed within it, yet not to be distinguished microscopically from the harmless germs of *Bacillus subtilis*. The bacterium of splenic fever is called *Bacillus Anthracis*. This formidable organism was shown to me by M. Pasteur in Paris last July. His recent investigations regarding the part it plays pathologically certainly rank amongst the most remarkable labours of that remarkable man. Observer after observer had strayed and fallen in this land of pitfalls, a multitude of opposing conclusions and mutually destructive theories being the result. In association with a younger physiological colleague, M. Joubert, Pasteur struck in amidst the chaos, and soon reduced it to harmony. They proved, among other things, that in cases where previous observers in France had supposed themselves to be dealing solely with splenic fever, another equally virulent factor was simultaneously active. Splenic fever was often overmastered by septicæmia, and results due solely to the latter had been frequently made the ground of pathological inferences regarding the character and cause of the former. Combining duly the two factors, all the previous irregularities disappeared, every result obtained receiving the fullest explanation. On studying the account of this masterly investigation, the words wherewith Pasteur himself feelingly alludes to the difficulties and dangers of the experimenter's art came home to me with especial force: 'J'ai tant de fois éprouvé que dans cet art difficile de l'expérimentation les plus habiles bronchent à chaque pas, et que l'interprétation des faits n'est pas moins périlleuse.'[1]

[1] Comptes-Rendus, lxxxiii. p. 177.

APPENDIX.

The following Abstract of Essay III., taken from the Proceedings of the Royal Society for 1877, *may be of use to the reader.*

For reasons which will appear in the sequel, it will be desirable to glance, in the first place, at the results already submitted to the Royal Society.

Portions of the autumn of 1875, and of the winter and spring of 1875–76, were devoted to the first section of these researches, and on the 13th of January, 1876, its main results were communicated orally to the Royal Society. The completed memoir was handed in to the Society on the 6th of April: it is published in vol. 166 of the 'Philosophical Transactions.'

Many of the 'closed chambers' employed in the inquiry were submitted on the 13th of January to the inspection of the Fellows. There had been over fifty of them in all, and several of them had been used more than once. The air in these chambers had been permitted to free itself from floating matter by self-subsidence, no artificial means of cleansing it being employed. Sterilized organic liquids and infusions of the most varied kinds freely exposed to air thus spontaneously purified were found, when tested by the microscope, to remain absolutely free from organisms of all kinds, and equally free from the turbidity, scum, and mould which to the naked eye are the infallible signs of the generation and multiplication of such organisms.

These experiments embraced, among others, the follow-

ing organic liquids :—urine in its natural conditions; infusions of mutton, beef, pork, hay, turnip, sole, haddock, codfish, salmon, turbot, mullet, herring, eel, oyster, whiting, liver, kidney, hare, rabbit, barndoor fowl, pheasant and grouse.

The number of separate vessels containing these liquids which were exposed to spontaneously purified air amounted to several hundreds, and the consensus of their testimony, in the sense just indicated, was complete.

Five minutes' boiling was found in all cases sufficient to sterilize the infusions.

When, after remaining sterile for months, the doors of the chambers were opened so as to admit the uncleansed air of the laboratory, the contact of such air, or, more correctly, of the matter mechanically floating in it, infallibly produced organisms in abundance—sometimes exclusively Bacterial, sometimes exclusively fungoid, and sometimes a combination of both.

Infusions of the substances above referred to were afterwards exposed in succession to air which had been freed of its floating matter by filtration through cotton wool, also to air from which the floating matter had been removed by calcination, and finally to vacua obtained by exhausting as far as possible with an air-pump, large receivers which had been previously filled with filtered air.

Boiled for five minutes and exposed to air thus treated, or to vacua thus produced, none of the infusions showed subsequently any alteration of colour or of transparency to the naked eye, or to the microscope any manifestation of life.

Thus far are summed up the results obtained with self-purified air, filtered air, calcined air, and air-pump vacua, the liquids in all cases being exposed in open test-tubes. Small retort-flasks, with drawn-out necks, were afterwards resorted to. Charged with the infusions, they were boiled in heated oil or brine, and sealed with exceeding care during ebullition. At the Royal Society on January 13, 1876, one hundred and thirty such flasks were submitted to the Fellows, free alike from putrefaction

and from life. They embraced specimens of all the substances above mentioned and some others.

Briefly expressed, then, the evidence furnished by six months' assiduous work during the autumn, winter, and spring of 1875–76, proved conclusively that in the atmospheric conditions then existing in the laboratory of the Royal Institution, not one of the many hundred flasks and tubes experimented on failed to be sterilized by five minutes' boiling, and no countenance was given to the notion that any of these once sterilized infusions possessed the power of spontaneously generating life.

The investigation embodied in the memoir now submitted to the Society was opened in the summer of 1876 by a series of tentative experiments on turnip-infusions, to which were added varying quantities of bruised or pounded cheese. Seven different kinds of cheese were employed, fifty-seven test-tubes being charged with the mixture and exposed to the self-purified air of closed chambers.

The majority of these mixtures remained unchanged; a minority became charged with organisms, which are, in my opinion, completely accounted for by reference to the protective action of the cheese. In the memoir of which this is an abstract such protective action is illustrated by the fact that when ordinary mustard seeds were tied together in a calico bag, they resisted the boiling temperature for a considerable multiple of the time which sufficed to kill them when no bag enveloped them. The bag and outside seeds protected the interior ones.

Not temperature alone, but the ability to diffuse its juices or salts, is a condition of prime importance in the destruction of the integrity and life of a germ by boiling water. Without diffusion a germ may withstand temperatures competent to utterly destroy it where diffusion is free. I need not remark on the imperviousness of cheese to water, and its consequent power to prevent diffusion.

These summer experiments on turnip-cheese infusions were, however, merely tentative, and I purpose completing them hereafter.

In the autumn I resumed experiments on infusions of hay, which had been purposely postponed. With this substance no difficulty was encountered in my first inquiry. Boiled for five minutes, and exposed to air purified spontaneously, or freed from its floating matter by calcination or filtration, hay-infusion, though employed in multiplied experiments at various times, never showed the least competence to kindle into life. After months of transparency, in a great number of cases, I inoculated this infusion with specks of animal and vegetable liquids containing *Bacteria*, and observed, twenty-four hours afterwards, its colour lightened and its mass rendered opaque by the multiplication of these organisms.

But in the autumn of 1876, the substance with which I had experimented so easily and successfully a year previously appeared to have changed its nature. The infusions extracted from it bore, in some cases, not only five minutes' but fifteen minutes' boiling with impunity. On changing the hay a different result was often obtained. Many of the infusions extracted from samples of hay purchased in the autumn of 1876 behaved exactly like those extracted from the hay of 1875, being completely sterilized by five minutes' boiling.

The possible influence of age and dryness soon suggested itself, and I tested the surmise to the uttermost. Numerous and laborious experiments were executed with hay derived from different localities; and by this means, in the earlier days of the inquiry, it was revealed that the infusions which manifested this previously unobserved resistance to sterilization were, one and all, extracted from old hay, while the readily sterilized infusions were extracted from new hay, the germs adhering to which had not been subjected to long-continued desiccation.

As the inquiry proceeded the distinction between old and new hay became more and more blurred, while prolonged experiment with hay of various kinds failed to rescue the question from uncertainty. I therefore turned to substances of a succulent nature—to fungi, cucumber,

melon, beetroot and artichoke, for example, whose parasitic or epiphytic germs were unlikely to have suffered desiccation.

Boiled for periods varying from five to fifteen minutes and exposed afterwards to moteless air, in numberless experiments these infusions broke down, charging themselves throughout with organisms, and loading themselves, almost in all cases, with a soapy corrugated scum.

I then fell back upon infusions whose deportment had been previously familiar to me, and in the sterilization of which I had never experienced any difficulty. Fish, flesh, and vegetables were re-subjected to trial. Though the precautions taken to avoid contamination were far more stringent than those observed in my first inquiry, and though the interval of boiling was sometimes tripled in duration, these infusions, in almost every instance, broke down. Spontaneously purified air, filtered air, and calcined air (calcined, I may add, with far greater severity than was found necessary a year previously) failed, in almost all cases, to protect the infusions from putrefaction.

I was sometimes cheered by a success which, at the time of its occurrence, would seem to be the result of increased severity in the methods of experiment. But the success was subsequently so opposed by failure that it finally stood out rather as an accident than as the normal result of the inquiry.

I had the most implicit confidence in the correctness of my earlier experiments; indeed incorrectness would have led to consequences exactly opposite to those arrived at. Errors of manipulation would have filled my tubes and flasks with organisms, instead of leaving them transparent and void of life. By the unsuccessful experiments above referred to a clear issue was therefore raised:—Either infusions of fish, flesh and vegetables had become endowed in 1876 with an inherent generative energy which they did not possess in 1875, or some new contagium external to the infusions, and of a far more obstinate character than that of 1875, had been brought to bear upon them at the later

date. The scientific mind will not halt in its decision between these two alternatives.

For my own part the gradual but irresistible interaction of thought and experiment made it in the first instance probable, and at last certain, that the atmosphere in which I worked had become so virulently infective as to render utterly impotent precautions against contamination and modes of sterilization which had been found uniformly successful in a less contaminated air. I therefore removed from the laboratory, first to the top, and afterwards to the basement of the Royal Institution, but found that even here, in a multitude of cases, failure was predominant, if not uniform. This hard discipline of defeat was needed to render me acquainted with all the possibilities of infection involved in the construction of my chambers and the treatment of my infusions.

I finally resolved to break away from the Royal Institution, and to seek at a distance from it a less infective atmosphere. In Kew Gardens, thanks to the President of the Royal Society, the requisite conditions were found. I chose for exposure in the Jodrell laboratory the special infusions which had proved most intractable in the laboratory of the Royal Institution. The result was that liquids which in Albemarle Street resisted two hundred minutes' boiling, becoming afterwards crowded with organisms, were utterly sterilized by five minutes' boiling at Kew.

A second clear issue is thus placed before the Royal Society:—Either the infusions had lost in Kew Gardens an inherent generative energy which they possessed in our laboratory, or the remarkable instances of life-development, after long-continued boiling, observed in the laboratory are to be referred to the contagium contained in its vessels or diffused in its air.

With a view to making nearer home experiments similar to those executed at Kew, I had a shed erected on the roof of the Royal Institution. In this shed infusions were prepared and introduced into new chambers of burnished tin, which had never been permitted to enter our labora-

tory. After their introduction the liquids were boiled for five minutes in an oil-bath.

The first experiment in this shed resulted in complete failure. Not one of the infusions exposed to the moteless air of the shed escaped putrefaction.

Either of two causes, or both of them combined, might, from my point of view, have produced this result. First, a flue from the laboratory was in free communication with the atmosphere not far from the shed; secondly, and this was the real cause of the infection, my assistants, in preparing the infusions, had freely passed from the laboratory to the shed. They had thus incautiously carried the contagium by a mode of transfer known to every physician.

The infected shed was disinfected; the infusions were again prepared, and care was taken, by the use of proper clothes, to avoid the former causes of contamination. The result was similar to that obtained at Kew, viz. organic liquids which, in the laboratory, withstood two hundred minutes' boiling, were rendered permanently barren by five minutes' boiling in the shed.

A third clear issue is thus placed before us which I should hardly think of formulating were it not for the incredible confusion which apparently besets this subject in the public mind. A rod thirty feet in length would stretch from the infusions in the shed to the same infusions in the laboratory. At one end of this rod the infusions were sterilized by five minutes' boiling, at the other end they withstood two hundred minutes' boiling. As before, the choice rests between two inferences:— Either we infer that at one end of the rod animal and vegetable infusions possess a generative power which at the other end they do not possess, or we are driven to the conclusion that at the one end of the rod we have infective and at the other end uninfective air.

The second inference is that which will be accepted by the scientific mind. To what, then, is the inferred difference at the two ends of the rod to be ascribed? In one obvious particular the laboratory this year differed from

that in which my first experiments were made. On its floor were various bundles of old and desiccated hay, from which, when stirred, clouds of fine dust ascended into the atmosphere. This dust proved to be both fruitful and, in the highest degree, resistant. Prior to the introduction of the hay which produced the dust, no difficulty as regards sterilization had ever been experienced; subsequent to its introduction my difficulties and defeats began.

I have twice glanced at periods of boiling amounting to two hundred minutes; for, after long and laborious trials of shorter periods, I advanced to longer ones, subjecting turnip, cucumber, and other infusions to the boiling temperature for intervals varying from five minutes to three hundred and sixty minutes. Up to a certain point these liquids maintained their power of developing life, but beyond this point complete sterility was the result. In the preliminary experiments bearing upon this question the point of sterilization lay between 180 and 240 minutes. Boiled for the former period the infusions continued fruitful; boiled for the latter period they remained permanently barren.

In these and numerous other experiments a method was followed which had been substantially employed by Spallanzani and Needham, and more recently by Wyman and Roberts, the method having been greatly refined by the philosopher last named. The flasks were partially filled with the infusions, the portions unoccupied by the liquids being taken up with ordinary unfiltered air. Now as regards the death-point of contagia we know that in air it may be much higher than in water, the selfsame temperature being fatal in the latter and sensibly harmless in the former; hence the doubt whether, in my recent experiments, the resistance of the contagium did not arise from the fact of its not being surrounded by liquid water.

I changed the method, and made a long series of experiments with filtered air. They were almost as unsuccessful as those made with ordinary air. From time to time I succeeded in producing complete sterility by five minutes' boiling; but these successes were so checked by

failures that, similar to other cases referred to, they finally appeared in the light of accidents. They were, however, by no means uninstructive, for they revealed the existence of breaks in the prevalence of the contagium, which, under the circumstances, might have been foreseen.

A rapid glance at the means employed to improve the methods of experiment, and at the results of their employment, may be permitted here. Bulbs, exhausted by an air-pump and afterwards heated almost to redness, were filled when cool with filtered air. While being charged with the infusions the bulbs were warmed, so as to produce a gentle outflow of air, and their necks were sealed while the outflow continued. It was thus sought to avoid the contamination consequent on an indraught.

The failures resulting from this mode of experiment greatly predominated over the successes.

Employing similar bulbs, their necks in the first instance were drawn out at the ends to tubes of capillary fineness. The bulbs were then filled each with one-third of an atmosphere of filtered air, and, while connected with the air-pump, were heated almost to redness. The capillary tubes were then sealed with the lamp. The sealed ends were afterwards broken off in the body of the liquid, two-thirds of each bulb being thus filled with the infusion. By great care it was found possible to re-seal the capillary tubes without removing them from the liquid. The infusions were afterwards boiled from five to fifteen minutes.

Here also the fruitfulness of the boiled infusion was the rule, and its barrenness the exception.

A source of discomfort clung persistently to my mind throughout these severer experiments. I was by no means certain that the observed development of life was not due to germs entangled in the film of liquid adherent to the necks and higher interior surfaces of the bulbs. This film might have evaporated, and its germs, surrounded by air and vapour, instead of by water, might, on this account, have been able to withstand an ordeal to which they would have succumbed if submerged.

A plan was therefore resorted to, by which the infusions were driven by atmospheric pressure through lateral channels issuing from the centres of the bulbs. As before, each bulb was filled with one-third of an atmosphere of filtered air, and afterwards heated nearly to redness. When fully charged, the infusion rose higher than the central orifice, and no portion of the internal surface was wetted save that against which the liquid permanently rested. The lateral channel was then closed with a lamp without an instant's contact being permitted to occur between any part of the infusion and the external air. It was thus rendered absolutely certain that the contagia exposed subsequently to the action of heat were to be sought, neither in the superjacent air nor on the interior surfaces of the flasks, but in the body of the infusions themselves.

By this method I tested in the first place the substance which, at an early stage of the inquiry, had excited my suspicion—without reference to which the discrepancy between the behaviour of infusions examined in the winter of 1875–76 and those examined in the winter of 1876–77 is inexplicable, but by reference to which the explanation of the observed discrepancy is complete ; I mean, the old hay which cumbered our laboratory floor.

Four hours' continuous boiling failed to sterilize bulbs charged with infusions of this hay. In special cases, moreover, germs were found so indurated and resistant that five, six, and, in one case, even eight hours' boiling failed to deprive them of life.

All the difficulties encountered in this long and laborious inquiry were traced to the germs which exhibited the extraordinary powers of resistance here described. They introduced a plague into our atmosphere—the other infusions, those of fresh hay included, like a smitten population, becoming the victims of a contagium foreign to themselves.[1]

It is a question of obvious interest to the scientific

[1] A hard and wiry hay from Guildford, which I have no reason to consider old, was found extremely difficult to sterilize.

surgeon whether those powerfully resistant germs are amenable to the ordinary processes of disinfection. It is perfectly certain that they resist to an extraordinary extent the action of heat. How would they behave in the wards of an hospital? There are, moreover, establishments devoted to the preserving of meats and vegetables. Do they ever experience inexplicable reverses? I think it certain that the mere shaking of a bunch of desiccated hay in the air of an establishment of this character might render the ordinary process of boiling for a few minutes utterly nugatory, thus possibly entailing serious loss. They have, as will subsequently appear, one great safeguard in the complete purgation of their sealed tins of air.

Keeping these germs, and the phases through which they pass to reach the developed organism, clearly in view, I have been able to sterilize the most obstinate infusions encountered in this inquiry, by heating them for a small fraction of the time above referred to as *insufficient* to sterilize them. The fully developed *Bacterium* is demonstrably killed by a temperature of 140° F. Fixing the mind's eye upon the germ during its passage from the hard and resistant to the plastic and sensitive state, it will appear in the highest degree probable that the plastic stage will be reached by different germs in different times. Some are more indurated than others, and require a longer immersion to soften and germinate. For all known germs there exists a period of incubation, during which they prepare themselves for emergence as the finished organisms which have been proved so sensitive to heat. If during this period, and well within it, the infusion be boiled for even the fraction of a minute, the softened germs which are then approaching their phase of final development will be destroyed. Repeating the process of heating every ten or twelve hours, before the least *sensible* change has occurred in the infusions, each successive heating will destroy the germs then softened, until, after a sufficient number of heatings, the last living germ will disappear.

Guided by the principle here laid down, and applying the heat discontinuously, infusions have been sterilized by an aggregate period of heating, which, fifty times multiplied, would fail to sterilize them if applied continuously. Four minutes in the one case can accomplish what four hours fail to accomplish in the other.

If properly followed out, the method of sterilization here described is infallible. A temperature, moreover, far below the boiling-point suffices for sterilization.

Another mode of sterilization, equally certain and remarkable, was forced upon me, so to speak, in the following way. In a multitude of cases a thick and folded layer of fatty scum, made up of matted *Bacteria*, gathered upon the surfaces of the infusions, the liquid underneath becoming sometimes cloudy throughout, but frequently maintaining a transparency equal to that of distilled water. The living scum-layer, as Pasteur has shown in other cases, appeared to possess the power of completely intercepting the atmospheric oxygen, appropriating the gas and depriving the germs in the liquid underneath of an element necessary to their development.

Placing the infusions in flasks, with large air-spaces above the liquids, corking the flasks, and exposing them for a few days to a temperature of 80° or 90° F., at the end of this time the oxygen of the superjacent air seems completely consumed. A lighted taper plunged into the flask is immediately extinguished. Above the scum, moreover, the interior surfaces of the bulbs used in my experiments were commonly moistened by the water of condensation. Into it the *Bacteria* sometimes rose, forming a kind of gauzy film to a height of an inch or more above the liquid. In fact, wherever air was to be found, these *Bacteria* followed it. It seemed a necessity of their existence. Hence the question, What will occur when the infusions are deprived of air?

I was by no means entitled to rest satisfied with inference as an answer to this question; for Pasteur has

abundantly demonstrated that the process of alcoholic fermentation depends on the continuance of life without air—other organisms than *Torula* being also alleged to be competent to live without oxygen. Experiment alone could determine the effect of exhaustion upon the particular organisms here under review.

Air-pump vacua were first employed, and with a considerable measure of success. Life was demonstrably enfeebled in such vacua.

Sprengel pumps were afterwards used to remove more effectually both the air dissolved in the infusions and that diffused in the spaces above them. The periods of exhaustion varied from one to eight hours, and the results of the experiments may be thus summed up :—Could the air be completely removed from the infusions, there is every reason to believe that sterilization *without boiling* would in most, if not in all, cases be the result. But, passing from probabilities to certainties, it is a proved fact that in numerous cases unboiled infusions deprived of air by five or six hours' action of the Sprengel pump are reduced to permanent barrenness. In a great number of cases, moreover, where the unboiled infusion would have become cloudy, exposure to the boiling temperature for a single minute sufficed completely to destroy the life already on the point of being extinguished through defect of air. With a single exception, I am not sure that any infusion escaped sterilization by five minutes' boiling after it had been deprived of air by the Sprengel pump. These five minutes accomplished what five hours sometimes failed to accomplish in the presence of air.

The exception here referred to is old-hay infusion, which, though sterilized in less than half the time needed to kill its germs where air is present, maintained a power of developing a feeble but distinct life after having been boiled for a large multiple of the time found sufficient to render infusions of mutton, beef, pork, cucumber, turnip, beetroot, shaddock, and artichoke permanently barren.

These experiments gave me the clue to many others

which might have readily become subjects of permanent misinterpretation. In the midst of a most virulently infective atmosphere, where, even after some hours' boiling, there was no escape for infusions supplied with air, the expulsion of the air by less than five minutes' boiling in properly shaped retort-flasks, and the proper sealing of the flasks during ebullition, insured the sterility of the infusions.

The meaning of a former remark regarding the part played by boiling, in establishments devoted to the preserving of meats and vegetables, will be now understood.

The inertness of the germs in liquids deprived of air is not due to a mere *suspension* of their powers. The germs are *killed* by being deprived of oxygen. For when the air which has been removed by the Sprengel pump is, after some time, carefully restored to the infusion, unaccompanied by germs from without, there is no revival of life. By removing the air we stifle the life which the returning air is incompetent to restore.

These experiments on the mortality arising from a defect of oxygen are, in a certain sense, complementary to the beautiful results of M. Paul Bert. Applying his method to my infusions, I find them sterilized in oxygen possessing a pressure of ten atmospheres or more. Like higher organisms, our Bacterial germs are poisoned by the excess and asphyxied by the defect of oxygen.

A few short sections on *Bacteria* germs as distinguished from *Bacteria* themselves,[1] and on the alleged destruction of germs by merely drying them, on hermetic sealing, and on the deportment of hermetically-sealed flasks exposed to the sun of the Alps, are introduced towards the end of the memoir.

[1] By the excellent researches of Dallinger and Drysdale it has been proved that the germs of Monads, as compared with the adult organisms, possess a power of resistance to heat in the proportion of 11 to 6.

NOTE ON THE DEPORTMENT OF ALKALIZED URINE.[1]

THE communication 'On the Influence of Liquor Potassæ and an Elevated Temperature on the Origin and Growth of Microphytes,' which, at Dr. Roberts's request, I have had the pleasure of presenting to the Royal Society, causes me to say earlier than I should otherwise have done that the subject which has occupied Dr. Roberts's attention has also occupied mine, and that my results are identical with his.

In some of the experiments the procedure described by Dr. Roberts was accurately pursued, save in one particular which has reference to temperature. Small tubes with their ends finely drawn out were charged with a definite amount of caustic potash, and subjected for a quarter of an hour to a temperature of 220° Fahr. They were then introduced into flasks containing measured quantities of urine. The urine being boiled for five minutes, the flasks were hermetically sealed during ebullition. They were subsequently permitted to remain in a warm place sufficiently long to prove that the urine had been perfectly sterilized by the boiling. The flasks were then rudely shaken, so as to break the capillary ends of the potash-tubes and permit the liquor potassæ to mingle with the acid liquid. The urine thus neutralized was subsequently exposed to a constant temperature of 122° Fahr., which is pronounced by Dr. Bastian to be specially potent as regards the generation of organisms.

I have not found this to be the case; for ten flasks,

[1] From the Proceedings of the Royal Society, No. 176, 1876.

prepared as above described towards the end of last September, remained for more than two months perfectly sterile. I have no doubt that they would have remained so indefinitely.

Three retorts, moreover, similar to those employed by Dr. Bastian, and provided with potash-tubes, had fresh urine boiled in them on the 29th of September, the retorts being sealed during ebullition. Several days subsequently, the potash-tubes were broken and the urine neutralized. Subjected for more than two months to a temperature of 122° Fahr. they failed to show any signs of life.

These results are quite in accordance with those obtained by Dr. Roberts. His potash-tubes, however, were exposed to a temperature of 280° Fahr., while mine were subjected to a temperature of 220° only.

With regard to the raising of the potash to a temperature higher than that of boiling water, M. Pasteur is in advance both of Dr. Roberts and myself. In a communication to the French Academy, on the 17th of last July, M. Pasteur showed that when due care is taken to add nothing but potash (heated to redness if solid, or to 110° C. if liquid) to sterilized urine, no life is ever developed as a consequence of the alkalization.[1]

M. Pasteur has quite recently favoured me with sketches of the simple but effectual apparatus by means of which he has tested the conclusions of Dr. Bastian. Since his return from his vacation at Arbois, he has carefully gone over this ground with results, he reports to me, not favourable to Dr. Bastian's views.

I may add that I have by no means confined myself to the thirteen samples of urine here referred to. The experiments have already extended to one hundred and five instances, not one of which shows the least countenance to the doctrine of spontaneous generation.

[1] That alkaline liquids are more difficult to sterilize than acid ones was announced by Pasteur more than fourteen years ago. See 'Annales de Chimie,' 1862, vol. lxiv. p. 62.

The method of 'Discontinuous Heating' was first described in the following letter to Professor Huxley.[1]

Royal Institution, Feb. 14, 1877.

My dear Huxley,—In my 'Preliminary Note,' communicated to the Royal Society on the 18th of January, various infusions were referred to as manifesting an astonishing resistance to sterilization by heat. This resistance was traced to its source ; and I have been since informed that you were good enough to express at the time a very favourable opinion as to the significance and value of the results indicated.

It will, I think, now interest you to learn that the most obstinate of the infusions referred to in the 'Note' have been since rendered tractable by the application of very simple means. Following up the plain suggestions of the germ theory, I have been able, even in the midst of a virulently infective atmosphere, to sterilize all the infusions by a temperature lower than that of boiling water.

It is known that the prolonged application of a low temperature is often equivalent to the brief application of a higher one ; and you may therefore be disposed to conclude that in the experiments here referred to I have substituted time for intensity. This, however, is not the case. The result depends solely upon the manner in which the heat is applied. For example, I boil an infusion for fifteen minutes, expose it to a temperature of 90° Fahr., and find it twenty-four hours afterwards swarming with life. I submit a second sample of the same infusion to a temperature lower than that of boiling water for five minutes, and it is rendered permanently barren.

The secret of success here is an open one. I have already referred to the period of latency which precedes the clouding of infusions with visible *Bacteria*. During

[1] From the Proceedings of the Royal Society, No. 178, 1877.

this period the germs are being prepared for their emergence into the finished organism. They reach the end of this period of preparation successively—the period of latency of any germ depending upon its condition as regards dryness and induration. This, then, is my mode of proceeding:—Before the latent period of any of the germs has been completed (say a few hours after the preparation of the infusion), I subject it for a brief interval to a temperature which may be under that of boiling water. Such softened and vivified germs as are on the point of passing into active life are thereby killed; others not yet softened remain intact. I repeat this process well within the interval necessary for the most advanced of those others to finish their period of latency. The number of undestroyed germs is further diminished by this second heating. After a number of repetitions, which varies with the character of the germs, the infusion, however obstinate, is completely sterilized.

The periods of heating need not exceed a fraction of a minute in duration. Sum them up in the case of an infusion which they have perfectly sterilized; they amount altogether to, say, five minutes. Boil another sample of the same infusion continuously for fifteen or even sixty minutes, you fail to sterilize it, although the temperature is higher and its time of application more than tenfold that which, discontinuously applied, infallibly produces barrenness.

In a few weeks I hope to bring this entire subject under the notice of the Royal Society; meanwhile, if you think it would interest them, I should be glad if you would communicate to the Fellows this general statement of the most recent results of experiment.

Believe me,

Ever faithfully yours,

JOHN TYNDALL.

T. H. Huxley, Esq., Sec. R.S.

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